

State of Hawaii  
DEPARTMENT OF LAND AND NATURAL RESOURCES  
Division of Aquatic Resources  
Honolulu, Hawaii 96813

August 12, 2010

Board of Land  
and Natural Resources  
Honolulu, Hawaii

Request for Authorization and Approval to Issue a Papahānaumokuākea Marine National  
Monument Research Permit to Dr. Charles Littnan, National Marine Fisheries Service,  
Pacific Islands Fisheries Science Center, for Access to State Waters to Conduct Juvenile  
Hawaiian Monk Seal Survival-Enhancement Activities

The Division of Aquatic Resources (DAR) hereby submits a request for your authorization and approval for issuance of a Papahānaumokuākea Marine National Monument research permit to Dr. Charles Littnan, Leader, Hawaiian Monk Seal Research Program, Pacific Islands Fisheries Science Center, pursuant § 187A-6, Hawaii Revised Statutes (HRS), chapter 13-60.5, Hawaii Administrative Rules (HAR), and all other applicable laws and regulations.

The research permit, as described below, would allow entry and research activities to occur in the Papahānaumokuākea Marine National Monument (Monument), including the NWHI State Marine Refuge and the waters (0-3 nautical miles) surrounding the following sites:

- Nihoa Island
- French Frigate Shoals
- Laysan Island
- Lisianski Island, Neva Shoal
- Pearl and Hermes Atoll
- Kure Atoll

The activities covered under this permit would occur between August 1, 2010 and July 31, 2011.

The proposed activities are a renewal of work previously permitted and conducted in the Monument.

INTENDED ACTIVITIES

The applicant proposes efforts to help increase juvenile monk seal survival in the Northwestern Hawaiian Islands. These activities include

1. Feeding and treating prematurely weaned and other undernourished seals in a captive facility
2. Treating weaned/juvenile seals to decrease parasite loads

These activities focus on direct interventions with juvenile seals to improve their survival and thus better position the monk seal population for recovery in the future. Based on the applicant's past experience, scientific review, and detailed consultations with external specialists, these activities are among the general approaches identified as the interventions most likely to be successful.

Captive Feeding Program: The Hawaiian monk seal program would collect, as appropriate, juvenile seals (0-3 yrs old) to feed, treat, and protect in captivity. Selected seals would be those that are prematurely weaned, undernourished twins, or in such a condition that would otherwise perish.

Seals would be cared for in shore pens or transported to the Ford Island Research Facility in Honolulu with the intent to release them back to their natal site or Nihoa Island. This is dependent on the facility coming on-line. If that does not occur, animals would not be brought back to Honolulu.

Protocols for captive care activities are included in F-1c.

Worming Trial: Monk seals are known to host a variety of gastrointestinal parasites, and it has been noted that young seals infected with *Diphyllbothrium* spp. (tape worms) tend to be in poorer body condition than those uninfected. While parasites are likely not a primary cause of mortality in monk seals, they may further compromise animals already in ill health due to food limitation, thereby increasing their likelihood of dying.

The applicant proposes to conduct a study to evaluate the efficacy of anti-helminth treatment as a method to improve juvenile survival. This study would focus on seals at Laysan, Lisianski, and French Frigate Shoals. The objective would be to include all available candidate seals in the study, which are likely to be a maximum of 41, 29, and 47 seals on each island respectively. Candidate seals are those that fall in the target age class of 0-2 years.

All study subjects would be captured by hand and net, sedated, and feces collected for subsequent determination of parasite burden, measured, tagged if necessary, and given an oral dose of praziquantal (Droncit, Bayer) and fenbendazole (Panacur) or ivermectin (Ivomec). Seals would be recaptured at periodic intervals for morphological assessment (weight and measurements) and treatments, up to 4 times per year. Control seals would be handled and sampled in the same manner as treatment seals, but would only receive saline solution.

All interaction and take of Hawaiian monk seals would be in accordance with procedures delineated in permits issued by the NMFS Office of Protected Resources.

These activities would determine effective methods to aid in the recovery of endangered Hawaiian monk seals and thus maintain biodiversity within the Monument. As such, the activities directly support the Monument Management Plan's action plan 3.2.1 – Threatened and Endangered Species (through strategy TES-1: Support activities that advance recovery of the Hawaiian monk seal).

The activities described above may require the following regulated activities to occur in State waters:

- ☒ Removing, moving, taking, harvesting, possessing, injuring, disturbing, or damaging any living or nonliving monument resource
- ☒ Anchoring a vessel

### REVIEW PROCESS

The permit application was sent out for review and comment to the following scientific and cultural entities: Hawaii Division of Aquatic Resources, Hawaii Division of Forestry and Wildlife, Papahānaumokuākea Marine National Monument (NOAA/NOS), NOAA Pacific Islands Regional Office (NOAA-PIRO), United States Fish and Wildlife Service Hawaiian and Pacific Islands National Wildlife Refuge Complex Office, and the Office of Hawaiian Affairs (OHA). In addition, the permit application has been posted on the Monument Web site since June 21st, giving the public an opportunity to comment. The application was posted within 40 days of its receipt, in accordance with the Monument's Public Notification Policy.

### **Comments received from the scientific community are summarized as follows:**

Scientific reviews support the acceptance of this application.

The following concerns were raised. Applicant responses are noted below.

1. Has the Ford Island Captive Care Facility come on line? If not, when is it expected to be operational?
  - The applicant reports that the facility is projected to be ready in early 2011. Seals would not be brought to the main Hawaiian Islands unless and until an appropriate captive facility was available.
2. What is the survival rate of juvenile seals that are treated in a captive care facility once they are released back into the wild?
  - The applicant explains that past experience with survival after captive care is mixed, and that survival depends on many factors outside of human control. For example, several female seals that were cared for in captivity during the 1980's to mid-1990's are still alive today in the NWHI. These seals, in turn, have given birth to many of their own pups. On the other hand, historically, many seals have not survived long-term after release

from captivity. Several factors probably influence survival prospects, including the age and experience of seals, body condition of animals on release, and the conditions (food availability, competition, predation risk, etc.) at the release site. The intention is to conduct captive care and release operations in a way that will maximize benefit to the individuals involved and to the general population. More important than the absolute post-release survival rate is the difference in survival chances for seals with and without captive care. For example, if a seal needing care had no chance to live without intervention and could later be released with a 50% chance, that would be valuable. If however, survival chances could not be substantially increase, then intervention would not be justified. Consistent with this concept, captive care efforts focus on animals (e.g., prematurely weaned pups) whose circumstances afford them virtually no opportunity to survival without assistance.

**Comments received from the Native Hawaiian community are summarized as follows:**

Cultural reviews support the acceptance of this application. No concerns were raised.

**Comments received from the public are summarized as follows:**

No comments were received from the public on this application.

**Additional reviews and permit history:**

Are there other relevant/necessary permits or environmental reviews that have or will be issued with regard to this project? (e.g. MMPA, ESA, EA) Yes ☒ No ☐

If so, please list or explain:

- All activities are currently authorized under Scientific Research and Enhancement Permit No. 10137-03, issued by the Office of Protected Resources, National Marine Fisheries Service.
- Environmental Assessment on Issuance of a Permit for Field Research and Enhancement Activities on the Endangered Hawaiian Monk Seal resulted in a FONSI
- The Department has made an exemption determination for this permit in accordance chapter 343, HRS, and Chapter 11-200, HAR. See Attachment ("DECLARATION OF EXEMPTION FROM THE PREPARATION OF AN ENVIRONMENTAL ASSESSMENT UNDER THE AUTHORITY OF CHAPTER 343, HRS AND CHAPTER 11-200 HAR, FOR PAPA HANAUMOKU AKEA MARINE NATIONAL MONUMENT RESEARCH PERMIT TO CHARLES LITTNAN, NOAA FISHERIES, PACIFIC ISLANDS FISHERIES SCIENCE CENTER, FOR ACCESS TO STATE WATERS TO CONDUCT JUVENILE HAWAIIAN MONK SEAL SURVIVAL-ENHANCEMENT ACTIVITIES UNDER PERMIT PMNM-2010-055")

Has Applicant been granted a permit from the State in the past? Yes ☒ No ☐

If so, please summarize past permits:

- The Applicant has been granted several monk seal related Monument permits, including PMNM-2008-043 and PMNM-2009-030 for similar activities in 2008 and 2009.

Have there been any a) violations: Yes ☐ No ☒  
b) Late/incomplete post-activity reports: Yes ☒ No ☐

The applicant has completed and turned in a report for the 2009 portion of his previous permit (PMNM-2009-030), however the report for activities thus far in 2010 is still outstanding. The applicant is a frequent permittee who has never had a late report in the past, thus staff is confident that the remainder of his report will be received soon.

Are there any other relevant concerns from previous permits? Yes ☐ No ☒

#### STAFF OPINION

DAR staff is of the opinion that Applicant has properly demonstrated valid justifications for his application and should be allowed to enter the NWHI State waters and to conduct the activities therein as specified in the application with certain special instructions and conditions, which are in addition to the Papahānaumokuākea Marine National Monument Research Permit General Conditions. All suggested special conditions have been vetted through the legal counsel of the Co-Trustee agencies (see Recommendation section).

#### MONUMENT MANAGEMENT BOARD OPINION

The MMB is of the opinion that the Applicant has met the findings of Presidential Proclamation 8031 and this activity may be conducted subject to completion of all compliance requirements. The MMB concurs with the special conditions recommended by DAR staff.

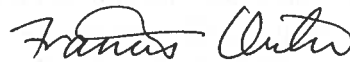
#### RECOMMENDATION

That the Board authorize and approve a Research Permit to Charles Littnan, Pacific Islands Fisheries Science Center, with the following special conditions:

1. This permit is not to be used for nor does it authorize the sale of collected organisms. Under this permit, the authorized activities must be for noncommercial purposes not involving the use or sale of any organism, by-products, or materials collected within the Monument for obtaining patent or intellectual property rights.

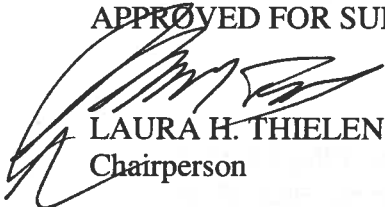
2. The permittee may not convey, transfer, or distribute, in any fashion (including, but not limited to, selling, trading, giving, or loaning) any coral, live rock, or organism collected under this permit without the express written permission of the Co-Trustees.
3. To prevent introduction of disease or the unintended transport of live organisms, the permittee must comply with the disease and transport protocol attached to this permit.
4. Tenders and small vessels must be equipped with engines that meet EPA emissions requirements.
5. Refueling of tenders and all small vessels must be done at the support ships and outside the confines of lagoons or near-shore waters in the State Marine Refuge.
6. No fishing is allowed in State Waters except as authorized under State law for subsistence, traditional and customary practices by Native Hawaiians.

Respectfully submitted,



 Administrator

APPROVED FOR SUBMITTAL



LAURA H. THIELEN  
Chairperson

LINDA LINGLE  
GOVERNOR OF HAWAII



STATE OF HAWAII  
DEPARTMENT OF LAND AND NATURAL RESOURCES  
DIVISION OF AQUATIC RESOURCES  
1151 PUNCHBOWL STREET, ROOM 330  
HONOLULU, HAWAII 96813

August 12, 2010

LAURA H. THIELEN  
CHAIRPERSON  
BOARD OF LAND AND NATURAL RESOURCES  
COMMISSION ON WATER RESOURCE MANAGEMENT


KEN C. KAWAHARA  
DEPUTY DIRECTOR - WATER

RUSSELL TSUJI  
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AQUATIC RESOURCES  
BOATING AND OCEAN RECREATION  
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FORESTRY AND WILDLIFE  
HISTORIC PRESERVATION  
KAHOOLAWE ISLAND RESERVE COMMISSION  
LAND  
STATE PARKS

TO: Division of Aquatic Resources File

THROUGH: Laura H. Thielen, Chairperson

FROM: Francis Oishi  
Division of Aquatic Resources 

DECLARATION OF EXEMPTION FROM THE PREPARATION OF AN ENVIRONMENTAL ASSESSMENT  
UNDER THE AUTHORITY OF CHAPTER 343, HRS AND CHAPTER 11-200 HAR, FOR  
PAPAHĀNAUMOKUĀKEA MARINE NATIONAL MONUMENT RESEARCH PERMIT TO CHARLES  
LITTNAN, NOAA FISHERIES, PACIFIC ISLANDS FISHERIES SCIENCE CENTER, FOR ACCESS TO STATE  
WATERS TO CONDUCT JUVENILE HAWAIIAN MONK SEAL SURVIVAL-ENHANCEMENT ACTIVITIES  
UNDER PERMIT PMNM-2010-055.

The following permitted activities are found to be exempted from preparation of an environmental assessment under the authority of Chapter 343, HRS and Chapter 11-200, HAR:

Project Title:

Papahānaumokuākea Marine National Monument Research Permit to Charles Littnan, NOAA Fisheries, Pacific Islands Fisheries Science Center, for Access to State Waters to Conduct Juvenile Hawaiian Monk Seal Survival-Enhancement Activities

Permit Number: PMNM-2010-055

Project Description:

The research permit application, as described below, would allow entry and activities to occur in Papahānaumokuākea Marine National Monument (Monument), including the NWHI State waters between August 1, 2010 through July 31, 2011.

This is an effort to help increase juvenile monk seal survival in the Northwestern Hawaiian Islands. Activities would include feeding and treating prematurely weaned and other undernourished seals in a captive facility, as well as treating weaned/juvenile seals with medication to decrease parasite loads.

The proposed activities are in direct support of the Monument Management Plan's priority management needs 3.2 – Conserving Wildlife and Habitats, through action plan 3.2.1 – Threatened and Endangered Species. This action plan includes a strategy to “support activities that advance recovery of the Hawaiian monk seal”.

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Activities to support conserving wildlife in the NWHI are addressed in the Monument Management Plan Environmental Assessment (December 2008) which resulted in a FONSI, or a finding of no significant impact. This EA specifically covers field activities, such as those being proposed, that will “increase juvenile survivorship through appropriate management tools, such as supplemental feeding through NOAA monk seal captive care programs” (PMNM MMP Vol 2, p.72). In addition, the “Environmental Assessment on Issuance of a Permit for Field Research and Enhancement Activities on the Endangered Hawaiian Monk Seal” resulted in a FONSI.

Consulted Parties:

The permit application was sent out for review and comment to the following scientific and cultural entities: Hawaii Division of Aquatic Resources, Hawaii Division of Forestry and Wildlife, Papahānaumokuākea Marine National Monument (NOAA/NOS), NOAA Pacific Islands Regional Office (NOAA-PIRO), United States Fish and Wildlife Service Hawaiian and Pacific Islands National Wildlife Refuge Complex Office, and the Office of Hawaiian Affairs (OHA). In addition, the permit application has been posted on the Monument Web site since June 21st, giving the public an opportunity to comment. The application was posted within 40 days of its receipt, in accordance with the Monument’s Public Notification Policy.

Exemption Determination:

After reviewing HAR § 11-200-8, including the criteria used to determine significance under HAR § 11-200-12, DLNR has concluded that the activities under this permit would have minimal or no significant effect on the environment and that issuance of the permit is categorically exempt from the requirement to prepare an environmental assessment based on the following analysis:

1. All activities associated with this permit, including captive care and deworming activities, have been evaluated as a single action. As a preliminary matter, multiple or phased actions, such as when a group of actions are part of a larger undertaking, or when an individual project is precedent to or represents a commitment to a larger project, must be grouped together and evaluated as a single action. HAR § 11-200-7. Since this permit involves an activity that is precedent to a later planned activity, i.e. the continuation of juvenile monk seal survival-enhancement activities, the categorical exemption determination here will treat all planned activities as a single action.

2. The Exemption Class for Scientific Research with no Serious or Major Environmental Disturbance Appears to Apply. Chapter 343, HRS, and § 11-200-8, HAR, provide for a list of classes of actions exempt from environmental assessment requirements. HAR §11-200-8.A.5. exempts the class of actions which involve “basic data collection, research, experimental management, and resource evaluation activities which do not result in a serious or major disturbance to an environmental resource.” This exemption class has been interpreted to include “wildlife surveys...recording, sampling” as well as “housing, care, feeding, veterinarian examination, breeding, cross fostering....of native species (including those which are rare, threatened, or endangered)”, such as those being proposed.

The proposed activities appear to fall squarely under the exemption class #5, exempt items #2 and #4 as described under the division of Forestry and Wildlife exemption list published on June 12, 2008. As discussed below, no significant disturbance to any environmental resource is anticipated in the sampling of Monument resources. Thus, so long as the below considerations are met, an exemption class should include the action now contemplated.



3. Cumulative Impacts of Actions in the Same Place and Impacts with Respect to the Potentially Particularly Sensitive Environment Will Not be Significant. Even where a categorical exemption appears to include a proposed action, the action cannot be declared exempt if “the cumulative impact of planned successive actions in the same place, over time, is significant, or when an action that is normally insignificant in its impact on the environment may be significant in a particularly sensitive environment.” HAR § 11-200-8.B. To gauge whether a significant impact or effect is probable, an exempting agency must consider every phase of a proposed action, any expected primary and secondary consequences, the long-term and short-term effects of the action, the overall and cumulative effect of the action, and the sum effects of an action on the quality of the environment. HAR § 11-200-12. Examples of actions which commonly have a significant effect on the environment are listed under HAR § 11-200-12.

The activities would be a continuation of work previously conducted by the applicant, which involved captive care and deworming activities to enhance juvenile monk seal survival. The applicant received a permit to conduct similar work in 2008 and 2009, and is likely to request future permits to continue this work. No deleterious effects have resulted from these activities in the past. No other studies of this type have been undertaken to date. With this in mind, significant cumulative impacts are not anticipated as a result of this activity, and numerous safeguards further ensure that the potentially sensitive environment of the project area will not be significantly affected. All activities would be conducted in a manner compatible with the management direction of the Monument Proclamation in that the activities do not diminish monument resources, qualities, and ecological integrity, or have any indirect, secondary, cultural, or cumulative effects. The joint permit review process did not reveal any anticipated indirect or cumulative impacts, nor did it raise any cultural concerns, that would occur as a result of these activities.

The activities would primarily be conducted by field biologists operating from the NOAA monk seal field camps throughout the Monument. Deployment of personnel and supplies at field camps throughout the Monument is covered under the Manager’s permit PMNM-2010-001, and is typically conducted from the NOAA Ship OSCAR ELTON SETTE which operates under permit PMNM-2010-006. Deworming activities would occur only at Laysan Island, and would be conducted throughout the year. Captive care collections could occur at any of the six main NWHI subpopulations areas (French Frigate Shoals, Laysan I., Lisianski I., Pearl and Hermes Reef, Midway Atoll and Kure Atoll). These activities would be conducted intermittently on a to-be-determined schedule based on the availability of vessels or other modes of transport.

One other permit, PMNM-2010-033, could potential take place in similar land/beach areas of the aforementioned islands and atolls concurrently. This permit, also issued to Charles Littnan, allows for the collection of GPS data within terrestrial areas of the Monument. Any other concurrent activities which involve Hawaiian monk seal interactions over the next year would also be coordinated and overseen by Charles Littnan’s team at the Pacific Islands Fisheries Science Center. As such, all potential concurrent activities would be highly coordinated and are not anticipated to have significant cumulative impacts.

Since no significant cumulative impacts or significant impacts with respect to any particularly sensitive aspect of the project area are anticipated, the categorical exemptions identified above should remain applicable.

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4. Overall Impacts will Probably be Minimal and Insignificant Any foreseeable impacts from the proposed activity will probably be minimal, and further mitigated by general and specific conditions attached to the permit. Specifically, all research activities covered by this permit will be carried out with strict safeguards for the natural, historic, and cultural resources of the Monument as required by Presidential Proclamation 8031, other applicable law and agency policies and standard operating procedures.

Conclusion. Upon consideration of the permit to be approved by the Board of Land and Natural Resources, the potential effects of the above listed project as provided by Chapter 343, HRS and Chapter 11-200 HAR, have been determined to be of probable minimal or no significant effect on the environment and exempt from the preparation of an environmental assessment.

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Laura H. Thielen  
Board of Land and Natural Resources

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Date

**Papahānaumokuākea Marine National Monument**  
**RESEARCH Permit Application**

***NOTE: This Permit Application (and associated Instructions) are to propose activities to be conducted in the Papahānaumokuākea Marine National Monument. The Co-Trustees are required to determine that issuing the requested permit is compatible with the findings of Presidential Proclamation 8031. Within this Application, provide all information that you believe will assist the Co-Trustees in determining how your proposed activities are compatible with the conservation and management of the natural, historic, and cultural resources of the Papahānaumokuākea Marine National Monument (Monument).***

**ADDITIONAL IMPORTANT INFORMATION:**

- Any or all of the information within this application may be posted to the Monument website informing the public on projects proposed to occur in the Monument.
- In addition to the permit application, the Applicant must either download the Monument Compliance Information Sheet from the Monument website OR request a hard copy from the Monument Permit Coordinator (contact information below). The Monument Compliance Information Sheet must be submitted to the Monument Permit Coordinator after initial application consultation.
- Issuance of a Monument permit is dependent upon the completion and review of the application and Compliance Information Sheet.

**INCOMPLETE APPLICATIONS WILL NOT BE CONSIDERED**

Send Permit Applications to:

Papahānaumokuākea Marine National Monument Permit Coordinator

6600 Kalaniana'ole Hwy. # 300

Honolulu, HI 96825

[nwhipermit@noaa.gov](mailto:nwhipermit@noaa.gov)

PHONE: (808) 397-2660      FAX: (808) 397-2662

**SUBMITTAL VIA ELECTRONIC MAIL IS PREFERRED BUT NOT REQUIRED. FOR ADDITIONAL SUBMITTAL INSTRUCTIONS, SEE THE LAST PAGE.**

## **Papahānaumokuākea Marine National Monument Permit Application Cover Sheet**

This Permit Application Cover Sheet is intended to provide summary information and status to the public on permit applications for activities proposed to be conducted in the Papahānaumokuākea Marine National Monument. While a permit application has been received, it has not been fully reviewed nor approved by the Monument Management Board to date. The Monument permit process also ensures that all environmental reviews are conducted prior to the issuance of a Monument permit.

### **Summary Information**

**Applicant Name:** Charles Littnan

**Affiliation:** NOAA Fisheries

**Permit Category:** Research

**Proposed Activity Dates:** August 1, 2010 - July 31, 2011

**Proposed Method of Entry (Vessel/Plane):** NOAA RV O.E. Sette

**Proposed Locations:** Kure Atoll, Midway Atoll, Pearl and Hermes Reef, Laysan, Lisianski, French Frigate Shoals, Nihoa

**Estimated number of individuals (including Applicant) to be covered under this permit:**

10

**Estimated number of days in the Monument:** 12 weeks

**Description of proposed activities: (complete these sentences):**

a.) The proposed activity would...  
consist of efforts to help increase juvenile monk seal survival in the Northwestern Hawaiian Islands.

b.) To accomplish this activity we would ....  
undertake enhancement actions including: 1) feeding and treating prematurely weaned and other undernourished seals in a captive facility, 2) treating weaned/juveniles to decrease parasite loads.

c.) This activity would help the Monument by ...  
determining effective methods to aid in the recovery of endangered Hawaiian monk seal and thus maintain biodiversity within the Monument.

**Other information or background:**

## **Section A - Applicant Information**

### **1. Applicant**

Name (last, first, middle initial): Littnan, Charles L.

Title: Leader, Hawaiian Monk Seal Research Program  
Pacific Islands Fisheries Science Center, NOAA Fisheries

#### **1a. Intended field Principal Investigator (See instructions for more information):**

Charles L. Littnan  
Kathleen Gobush

#### **2. Mailing address (street/P.O. box, city, state, country, zip):**

[REDACTED]

Phone:

[REDACTED]

Fax:

Email:

[REDACTED]

For students, major professor's name, telephone and email address:

#### **3. Affiliation (institution/agency/organization directly related to the proposed project):**

Pacific Islands Fisheries Science Center/NOAA Fisheries/Department of Commerce

#### **4. Additional persons to be covered by permit. List all personnel roles and names (if known at time of application) here (e.g. John Doe, Research Diver; Jane Doe, Field Technician):**

Tenaya Norris - Field Researcher  
Robert Braun - Veterinarian  
Frances Gulland - Veterinarian  
Chad Yoshinaga - Field Researcher  
Jessie Lopez - Field Researcher

Shawn Farry - Field Researcher  
TBD- additional field research staff

## **Section B: Project Information**

### **5a. Project location(s):**

<input checked="" type="checkbox"/> Nihoa Island	<input checked="" type="checkbox"/> Land-based	<input checked="" type="checkbox"/> Shallow water	<input type="checkbox"/> Deep water
<input type="checkbox"/> Necker Island (Mokumanamana)	<input type="checkbox"/> Land-based	<input type="checkbox"/> Shallow water	<input type="checkbox"/> Deep water
<input checked="" type="checkbox"/> French Frigate Shoals	<input checked="" type="checkbox"/> Land-based	<input checked="" type="checkbox"/> Shallow water	<input type="checkbox"/> Deep water
<input type="checkbox"/> Gardner Pinnacles	<input type="checkbox"/> Land-based	<input type="checkbox"/> Shallow water	<input type="checkbox"/> Deep water
<input type="checkbox"/> Maro Reef			
<input checked="" type="checkbox"/> Laysan Island	<input checked="" type="checkbox"/> Land-based	<input checked="" type="checkbox"/> Shallow water	<input type="checkbox"/> Deep water
<input checked="" type="checkbox"/> Lisianski Island, Neva Shoal	<input checked="" type="checkbox"/> Land-based	<input checked="" type="checkbox"/> Shallow water	<input type="checkbox"/> Deep water
<input checked="" type="checkbox"/> Pearl and Hermes Atoll	<input checked="" type="checkbox"/> Land-based	<input checked="" type="checkbox"/> Shallow water	<input type="checkbox"/> Deep water
<input checked="" type="checkbox"/> Midway Atoll	<input checked="" type="checkbox"/> Land-based	<input checked="" type="checkbox"/> Shallow water	<input type="checkbox"/> Deep water
<input checked="" type="checkbox"/> Kure Atoll	<input checked="" type="checkbox"/> Land-based	<input checked="" type="checkbox"/> Shallow water	<input type="checkbox"/> Deep water
<input type="checkbox"/> Other			

NOTE: There is a fee schedule for people visiting Midway Atoll National Wildlife Refuge via vessel and aircraft.

### **Location Description:**

Location will be the sand beaches and near shore habits of all islands specified, including sand beaches of all islets within the atoll complexes of Kure Atoll, Midway Atoll, Pearl & Hermes Atoll, and French Frigate Shoals. Most of the effort, however, will take place in these habitats at French Frigate Shoals, Laysan Island, Lisianski Island, and Nihoa Island.

### **5b. Check all applicable regulated activities proposed to be conducted in the Monument:**

- ☒ Removing, moving, taking, harvesting, possessing, injuring, disturbing, or damaging any living or nonliving Monument resource
- ☐ Drilling into, dredging, or otherwise altering the submerged lands other than by anchoring a vessel; or constructing, placing, or abandoning any structure, material, or other matter on the submerged lands
- ☒ Anchoring a vessel
- ☐ Deserting a vessel aground, at anchor, or adrift
- ☐ Discharging or depositing any material or matter into the Monument
- ☐ Touching coral, living or dead
- ☐ Possessing fishing gear except when stowed and not available for immediate use during passage without interruption through the Monument
- ☐ Attracting any living Monument resource
- ☐ Sustenance fishing (Federal waters only, outside of Special Preservation Areas, Ecological Reserves and Special Management Areas)
- ☐ Subsistence fishing (State waters only)
- ☐ Swimming, snorkeling, or closed or open circuit SCUBA diving within any Special Preservation Area or Midway Atoll Special Management Area

**6 Purpose/Need/Scope *State purpose of proposed activities:***

The Hawaiian monk seal is on a path to extinction that is unlikely to be altered without human intervention. This application presents tools for improving the survival of young seals, an essential requirement for averting extinction.

The total abundance of Hawaiian monk seals in the Northwestern Hawaiian Islands (NWHI), has declined by 70 % since the late 1950s. Since then, the six main sub-populations have experienced everything from periods of promising growth to catastrophic setbacks. The causes of decline have varied over time and from place to place, but since the early 1990s the decline has been driven, in large part, by poor juvenile survival. Many of these young animals have failed to thrive, and only about 1 of every 5 live to reach maturity, a situation largely due to insufficient food availability. The age structure of the population is therefore now unfavorable for future growth and the total population will inevitably fall below 1,000 individuals in just a few years.

The decline will continue and the conservation challenge will intensify unless scientists and managers, working together, develop the means to improve juvenile survival. History teaches us that the monk seal will continue to face new and unforeseen challenges in the future, but after two decades of poor juvenile survival, it is clear that this problem must be addressed. Improving juvenile survival is one of four key activities highlighted in the new Recovery Plan for the Hawaiian monk seal, published by NOAA in the summer of 2007:

- Improving juvenile survival through direct intervention such as providing captive care and feeding;
- Mitigating mortality due to entanglement in marine debris;
- Reducing shark predation on seal pups; and
- Ensuring growth of the small Main Hawaiian Islands seal population.

All of these critical recovery activities are being pursued by NOAA and its partners. The work proposed here focuses on direct interventions with juvenile seals to improve their survival and thus better position the monk seal population for recovery in the future.

**7. Answer the Findings below by providing information that you believe will assist the Co-Trustees in determining how your proposed activities are compatible with the conservation and management of the natural, historic, and cultural resources of the Monument:**

The Findings are as follows:

- a. How can the activity be conducted with adequate safeguards for the cultural, natural and historic resources and ecological integrity of the Monument?

The ultimate goal of the work described here is to assist in the recovery of the Hawaiian monk seal, a goal that is consistent with Monument mandates. The research proposed herein is



compatible with the conservation and management goals of the Monument and minimizes disturbance to the NWHI ecosystem.

Our studies will be designed and executed so as to minimize impacts to the terrestrial and marine environment. For instance, on-island time will be limited to that required for animal capture, transport, and instrument deployment, during which all personnel will adhere to strict quarantine protocols as defined by USFWS. Movements will be confined to the immediate beach area to avoid potential disturbance to bird and plant life on the island interiors. After the final tag deployment or adequate monitoring period, NMFS monk seal researchers will arrange to return to the NOAA R/V Oscar Elton Sette or other vessel, thereby reducing any human disturbance to terrestrial habitats and species by returning early.

Native Hawaiians share a close link to the ocean, marine life, and islands within the monument and seek to maintain the living cultural resources found there. Hawaiian monk seals are one of the most threatened of these cultural and natural legacies. The work presented here is critical for the survival of this species into the future, and it is our intent to continue this work with respect and in partnership with the Native Hawaiian community. Accordingly, all scientists participating on these cruises will receive a Native Hawaiian cultural briefing before departure to the NWHI. In addition, the primary permittee, chief scientist, and other appropriate personnel look forward to consulting with the Office of Hawaiian Affairs (OHA) and the Monument's Native Hawaiian program coordinator on proper conduct while in the NWHI, on cultural sensitivities associated with the proposed activities and locations, and on the applicability of the results of this research to the role of OHA as one of the NWHI stakeholder agencies.

b. How will the activity be conducted in a manner compatible with the management direction of this proclamation, considering the extent to which the conduct of the activity may diminish or enhance Monument cultural, natural and historic resources, qualities, and ecological integrity, any indirect, secondary, or cumulative effects of the activity, and the duration of such effects? Please see 7a.

c. Is there a practicable alternative to conducting the activity within the Monument? If not, explain why your activities must be conducted in the Monument.  
The techniques proposed here to improve juvenile survival can only be applied to seals in the NWHI. This population, unlike seals in the MHI, is demonstrating a population decline and nutritionally stressed seals.

d. How does the end value of the activity outweigh its adverse impacts on Monument cultural, natural and historic resources, qualities, and ecological integrity?  
The potential gain from this project is the increased survival of juvenile monk seals in the rapidly dwindling NWHI population. This work if successful and applied on a broader scale in the future could slow or stop the population decline.

e. Explain how the duration of the activity is no longer than necessary to achieve its stated purpose.

All activities here are devised in a manner to minimize time in the field. Researchers will remain in the field for only the time necessary to handle, treat and monitor seals to a degree that ensures the success of the studies and actions proposed here. Most work proposed here is also intended to occur in conjunction with population assessment camps already in place or replace additional foraging trips by using seals for multiple purposes (i.e. worming trial and foraging research).

f. Provide information demonstrating that you are qualified to conduct and complete the activity and mitigate any potential impacts resulting from its conduct.

The Hawaiian Monk Seal Research Program has been conducting research on this species for over two decades. All members participating on these studies have previous monk seal handling experience. The protocols and research plans presented for these studies have been reviewed and approved by a variety of experts including the Marine Mammal Commission, Hawaiian Monk Seal Recovery Team, as well as other external specialists.

g. Provide information demonstrating that you have adequate financial resources available to conduct and complete the activity and mitigate any potential impacts resulting from its conduct. All research/enhancement activities are supported by NOAA Fisheries funding and primarily with the use of NOAA research vessels.

h. Explain how your methods and procedures are appropriate to achieve the proposed activity's goals in relation to their impacts to Monument cultural, natural and historic resources, qualities, and ecological integrity.

All participating staff are educated and trained to respect all cultural, natural and historic resources in the Monument. Our first and primary objective is "Do no harm". See section 7a above for details.

i. Has your vessel has been outfitted with a mobile transceiver unit approved by OLE and complies with the requirements of Presidential Proclamation 8031?

Yes

j. Demonstrate that there are no other factors that would make the issuance of a permit for the activity inappropriate.

There are no factors, such as other permit violations, that should prevent the issuance of this permit. All activities are inline with Hawaiian Monk Seal Recovery Plan and relevant sections of the Monument Management Plan. We have yet to submit a final report from our 2009 permit for similar work as it is still active until May 31, 2010. We will submit a final report within 60 days of that date.

#### **8. Procedures/Methods:**

A range of prospective approaches for increasing juvenile survival have been identified, including:

- Bringing young animals into captivity for feeding and veterinary care, followed by release back into the wild
- Treatment of free-ranging young animals to reduce parasite loads

Based on past experience, scientific review, and detailed consultations with external specialists, these three general approaches have been identified as the interventions most likely to be successful. Some latitude is required in the application of these interventions because at any given time, the optimal approach will depend on a number of factors such as the relative survival among the different sites, the logistics of moving animals, the availability of favorable release sites and so on.

### Captive Feeding Program

The Hawaiian monk seal program will collect, as appropriate, juvenile seals (0-3 years old) to feed, treat, and protect in captivity. Seals selected for this work will be those that are prematurely weaned, undernourished twins, or otherwise in a condition that without captive care and supplemental feeding will perish. Seals will be cared for in shore pens or transported to and cared for at the Ford Island Research Facility in Honolulu, Hawaii with the intent to release them back at their natal site or Nihoa Island (see translocation section) in the NWHI. Captive care operations will be dependent on the Ford Island Facility coming on-line in early 2010. If this does not occur, then animals will not be brought back to Honolulu.

### On-site operations.

Field operations will be needed to assess, capture, and hold animals that would benefit from interventions to improve their survival. Assessment of individual seals is a routine element of ongoing annual studies. Capturing seals is more complicated because juvenile animals, in particular, may be absent from the islands for weeks at a time. Therefore, on-site holding is almost always required because capture cannot be reliably timed to coincide with the arrival and departure of a transport vessel or aircraft.

### Transport of animals from NWHI to MHI and return.

As much as possible, captive care operations will be supported by existing vessel and aircraft activity associated with establishing and retrieving annual field camps. In the past, the U.S. Coast Guard, U.S. Navy, and U.S. Air Force have provided additional assistance opportunistically, and similar arrangements will be sought to minimize transportation costs. In spite of such welcome help, additional chartering of both vessels and aircraft may be necessary.

### Release and post-release monitoring.

Releasing animals may require temporary holding facilities if a “soft release” method (i.e., gradual introduction to the release site) is used. Staff will be needed to provide care at the release site, as well as to release and monitor the animals’ acclimation. Monitoring will involve observational assessment to gauge animal condition and health, as well as tracking movement and foraging patterns using well-established tagging technology. In many, if not most, cases, releases will be timed to take advantage of personnel, equipment, and support from concurrent field studies.

Examples of protocols for captive care activities are included in Appendices 1-5.

### Worming

#### Objective:

Hawaiian monk seal abundance is declining due to low juvenile survival, which appears to be associated with food limitation and poor body condition. Monk seals are known to host a variety of gastrointestinal parasites (Dailey et al. 1988, 2004). Reif et al. (2006) reported that young seals infected with *Diphyllbothrium* spp. (tape worms) tended to be in poorer body condition than those uninfected, and proposed that “intervention strategies to reduce the gastrointestinal helminth burdens in immature animals should be considered as a conservation measure.” To date, no studies have been conducted to evaluate the efficacy of anti-helminth treatment as a method to improve juvenile survival. This study, then, is designed to test the hypothesis that temporarily relieving compromised young monk seals of their parasite burden will improve their chances of survival in a food limited environment. Specifically, we will determine the potential for enhanced survival of seals, aged young-of-the-year to 2-years old, following treatment to reduce gastro-intestinal parasite load.

#### Experimental design and protocols:

##### Selection of subject animals (pre-treatment assessment)

The study will focus on NWHI juvenile seals up to two years of age which corresponds to the age range exhibiting the lowest survival and which is primarily responsible for constraining population growth and recovery. Sample sizes will be limited by the number of juveniles available at each study site which match the specified selection criteria (see below), or by the capability of the field teams to identify, capture and treat the seals safely in the allotted time. The objective will be to include all available candidate seals in the study. Based on cohort numbers over the last 5 years, the maximum number of seals in each age class (0-2) that may be included in the study each year are: French Frigate Shoals: 47 seals; Laysan Island: 41 seals; and Lisianski Island: 29 seals. Standard population surveys will be conducted to identify potential study subjects. We will attempt to treat all animals that fall into the target age class. IF the opportunity to treat seals is limited in certain cases and in order to test the hypothesis above, we will prioritize sampling for animals that are most likely to be compromised by nutritional stress and parasites, but which are not moribund and unlikely to survive under any circumstances.

Pre-treatment assessments of health status and body condition will be based on visual inspection, supplemented by examination of digital photos. The monk seal program employs a suite of body condition indices to score seals as good/medium/thin/emaciated. These indices rely primarily on the relative visibility of: “pelvic girdle” (ischium, greater trochanter of the femur, ilium), ribs, point of the shoulder (scapulohumoral joint), peanut head (circumferential loss of mass around the neck and cranial shoulders), and vertebrae (dorsal spinous processes and transverse processes). Seals scored as “emaciated” often disappear from the population soon thereafter or before the start of the subsequent field season, and if it is felt that they are too compromised to treat without severe risk of mortality they will be excluded from this study.

##### Controls

Seals will be assigned to either a treatment or control group using either random assignment or alternated systematically as they are encountered. The objective will be to obtain an equivalent

number of seals in both treatment and control groups, matched as closely as possible in age, sex, body condition, and location. Sex matching is important because sex has been recognized to influence worm burden and its effects on the host in other mammals (Wilson and Moore 2002).

#### Location and Timing

Historically, juvenile survival has varied markedly both temporally and spatially (Baker and Thompson 2006). Further, the pattern of the relationship between pup condition (weaning girth) and survival also varies annually, apparently due to environmental stochasticity (Baker et al. 2007), predation intensity and other factors (Baker in press). Due to this high degree of variability, in order to detect and describe the effects of the treatment, the study will be conducted at multiple sites and years. Each year, Laysan Island will serve as one of the study sites, selected because there is a relatively large number of pups born annually and apparently a minimum of mortality factors other than food limitation (e.g., predation) to confound the results. Also, because it is a single island, all animals on the beach on any given day are available for treatment and observation (as opposed to the greater logistic difficulties at multi-islet atolls). Additional study sites include Lisianski Island and French Frigate Shoals. Lisianski is similar to Laysan in terms of known mortality causes and physiography. FFS may serve as an additional site if funding and logistics prove favorable to extending the study.

Seals will be handled for treatment or assessment (sampling or weighing) up to eight times each year: Spring (February/March), Summer (June-August), Autumn (October) and Winter (December). Seals age 1-2 will be treated during any of these four times. Pups of the year will not be treated during the summer session (first treatment during the fall session) to ensure that they have had ample exposure to parasites, unless it is confirmed that the pup has been weaned for at least 120 days, in which case they may be treated). The constraint is based on results from epidemiological sampling conducted 1998-2000 (n=54 for weaned pups), which indicated that all pups sampled more than 120 days post-weaning (n=15) tested positive for either cestodes or nematodes (Appendix A: Figure 1). Conversely, only one pup sampled at less than 75 days post-weaning (n=39) tested positive. Based on those findings and the timing of the monk seal's annual birth pulse (peak parturition in May and June; Johanos et al. 1994), fall sampling (October) of weaned pups will ensure that most pups will have had ample exposure to parasites through feeding and will be infected.

#### Treatment and Assessment Protocols

All study subjects will be captured by hand and net, feces collected for subsequent determination of parasite burden/presence (voided feces or fecal sample collected via fecal loop or digital extraction; stored in 10 % formalin), measured (axillary girth and dorsal standard length and weight), tagged if necessary, and given an oral dose of praziquantal (Droncit, Bayer) at 5 mg/kg and 10-50 mg/kg fenbendazole (Panacur) or 0.5 mg/kg ivermectin (Ivomec) and released. Oral dosing may be done either with a pill or paste form of the drugs, in accordance with the protocols developed for elephant seals at The Marine Mammal Center, and applied to monk seals during captive rearing studies at Midway Atoll in 2006-7. Control seals will be handled and sampled in the same manner as treatment seals. No sedation will be required for either treatment or control seals. However, the study may be facilitated by conducting it in conjunction with other research involving capture and handling of juvenile monk seals (e.g., foraging and health screen studies),

in which case treatments may involve sedation, biomedical sampling, and instrumentation. Post treatment condition and fecal egg counts will be determined by observing the seals, and, when possible, collecting multiple voided fecal samples from known individuals as part of our standard patrols. Because condition differences are most likely to be maximal at 2-6 weeks post deworming (F. Gulland, pers. obs.), we will attempt to recapture seals in both treatment and control groups for morphological assessment (weight and measurements) and parasite sampling at approximately 4 weeks post-treatment. We will attempt to deworm all treatment seals (same protocol as for initial treatment) during each subsequent field session, or at approximately 7-12 week intervals. Thus, each seal may be treated up to four times per year, except pups-of-the-year which will be limited to a maximum of three treatments (no summer treatment). The additional treatments serve to clear both the adult worms that survived the previous treatments (or acquired thereafter), as well as migrating larva that matured after the previous treatment. As with the initial treatment, control seals will be handled and measured in the same manner as treatment seals, but will receive only saline solution. Fecal samples will be collected from all handled seals. As stated previously, attempts Visual assessment of condition will be recorded on an ongoing basis throughout the study, using standard MMRP subjective body condition scoring (med/thin/emaciated with +/- designations) and scat samples will be collected and preserved for detection of parasites. Subsequent survival will be determined through visual re-identification during regular monk seal population assessment field research, (typically June through August), supplemented by observations made during the additional field sessions for this or other projects. The duration of the survival period will be dependent upon the timing of the initial field phase of the study relative to the assessment field season. That is, at those sites lacking a constant field presence, seals that are not observed in the subsequent field session may have died at anytime during the interim from the previous observation. The resolution of the survival assessment is therefore limited by the frequency of the field presence.

#### Evaluation:

The evaluation of treatment effects will address two questions:

1. Does treatment of young seals improve their physiological condition (weight, body condition ranking, and parasite load)?
2. Is improved physiological condition (#1) sufficient to increase survival under favorable environmental conditions?

The primary statistical analysis will consist of modeling survival (either with capture-recapture or logistic regression) of treatment and control animals to determine whether there is evidence that anti-helminth treatment improves survival. Other factors that influence survival (predation or other) will be treated as covariates. However, our ability to identify and quantify the underlying environmental factors that drive juvenile survival rates is limited.

Although survival is the ultimate measure, it is a function of a number of factors (physiological condition, environmental conditions, shark predation, entanglement, and other), and worming may not be sufficient to boost survival in all cases. Under severe environmental conditions, animals might show an improvement in condition but not survival. However, that same increase will be made to measure treatment and control seals between each treatment session under more

moderate environmental conditions. Consequently, another important analysis will be a comparison of body condition change in treated versus control animals. This assessment will use both quantitative measures (weight, girth, and length) and qualitative measures (MMRP categorical body condition ranking). It should be noted that we might observe different patterns in the effect of worming treatment on seals depending on their life stage. Young of the year are likely to lose weight regardless of treatment as they lose their excess blubber and learn to forage, in this case we would hope that treated seals would show a less precipitous decline in mass and condition than non-treated individuals. Seals in their second year of life would do well to maintain condition and demonstrate some weight gain. Those starting their third year of life could gain weight and condition relative to non-treated animals.

Another key analysis is the comparison of parasite loads in control versus treatment seals. Parasite load will be determined from fecal egg count data, treated as a categorical covariate. Fecal egg counts are not an exact measure of the number of worms present in the stomach and intestine of live animals, as egg production by female worms is influenced by host immunity and worm burden (Gulland and Fox 1992, Zhong and Dobson 1996; Aumont et al. 2003). Furthermore, egg release by tapeworms is intermittent (Reif et al. 2006), whereas nematode eggs are usually randomly scattered in the stools, even if introduced into the fecal stream above the lower colon at irregular intervals as much as several hours apart (Martin 1965; LeJambre et al. 2007). Evaluation of a variety of gastrointestinal nematodes in a number of species has shown that despite variability in fecundity, the fecal egg count is sufficiently repeatable within an individual to allow its use as a quantitative measure of worm burden, and to allow comparisons between individuals (Coltman et al. 1999). Reduction of fecal egg count is the most widely used method to assess the efficacy of anthelmintics against gastrointestinal strongyles (Cabaret and Berrag, 2004). Thus we will use the fecal egg count to evaluate worm burden, and will aim to collect 3 fecal samples per time point to be evaluated, and calculate the mean fecal egg count from these 3 samples to minimize error due to intermittent worm shedding. Based on these results, samples will be classified into one of three categories corresponding to low, medium or high infection. Parasite load in any dead animals collected during the study will be determined through an absolute worm count.

Due to the aforementioned volatility in annual survival rates of juvenile monk seals, it is anticipated that unless the treatment effect is very large, definitive statistical conclusions may not emerge from the first year of the study. However, should preliminary conclusions suggest that the treatment is associated with a beneficial effect on monk seal condition, the study may be scaled up (in terms of number of subjects and/or locations) during the subsequent field seasons. Also, if indications are that worming improves survival, we will adjust the ratio of treatment to controls so that more seals are treated. Conversely, if there is any indication that the welfare of the subjects has been compromised by the handling, the treatment or any other artifact of the study, MMRP will initiate consultation with the Monk Seal Health Care Working Group to evaluate the preliminary results.

**NOTE: If land or marine archeological activities are involved, contact the Monument Permit Coordinator at the address on the general application form before proceeding, as a**

**customized application will be needed. For more information, contact the Monument office on the first page of this application.**

**9a. Collection of specimens - collecting activities (would apply to any activity): organisms or objects (List of species, if applicable, attach additional sheets if necessary):**

Common name:  
Hawaiian monk seal

Scientific name:  
*Monachus schauinslandi*

# & size of specimens:  
Captive Care

An unknown number of seals, but likely less than 10, may be brought to the MHI for captive care. During this time they will be biomedically sampled multiple times to monitor their health and condition.

**Worming Trial**

Up to 90 juvenile seals (ages 1 month - 2 years) will be captured, sampled and treated for the parasite study. Of these 45 will be controls that will be sampled and 45 treatment seals will be sampled and treated with anti-helminth drugs. Up to 200 feces collected for subsequent determination of parasite burden (voided feces or fecal sample via fecal loop stored in 10 % formalin), Up to 200 measurements of morphometrics (axillary girth and dorsal standard length), 90 x 2 (per animal) blubber biopsies (approx. 0.6 cm diameter, 2-3 cm in length) may be collected, 90 blood samples (up to 90 mL) may be collected, 90 swabs x 5 orifices (anal, genital, mouth, nose, eye), 200 Scats opportunistically collected on beach, Up to 90 x 2 skin plugs from flipper tagging.

There is also the possibility of conducting necropsies on any dead seals found during research activities. The type and number of samples collected during necropsies varies depending on the condition of the carcass. A necropsy protocol that highlights the potential tissues that may be collected from dead monk seals can be provided upon request, though tissues could include: samples from all major organs, skin, muscle, blood, blubber, hair, bone and other.

Collection location:  
Captive Care

Potentially any of the 6 main sub-populations in the NWHI as this an opportunistic effort.

**Worming Trial**

Lisianski Island, Laysan Island, French Frigate Shoals

☒ Whole Organism ☒ Partial Organism

**9b. What will be done with the specimens after the project has ended?**



Samples will be analyzed in a timely basis upon return to Honolulu. All samples collected and not analyzed during this project (i.e. duplicate blubber for fatty acids, skin for genetics) will be stored at the PIFSC or Bishop Museum for future analysis.

**9c. Will the organisms be kept alive after collection?** ☒ Yes ☐ No

- General site/location for collections:

Seals could be collected from any of the major breeding populations. They will be held in shore pens or brought to the MHI to be held in pools in a captive care facility.

- Is it an open or closed system? ☒ Open ☐ Closed

- Is there an outfall? ☒ Yes ☐ No

- Will these organisms be housed with other organisms? If so, what are the other organisms?  
They may be kept with other monk seals.

- Will organisms be released?

Yes

**10. If applicable, how will the collected samples or specimens be transported out of the Monument?**

All samples collected within the monument will be transported out on the NOAA/RV OES. Blubber and other tissue samples will be stored in a liquid nitrogen dewar. Most tissues will be stored in ethanol. Skin plugs from monk seals and cetaceans may be stored in DMSO prior to freezing. Fecal samples are stored in buckets and later frozen on the vessel.

**11. Describe collaborative activities to share samples, reduce duplicative sampling, or duplicative research:**

Currently NOAA Fisheries is the only group researching Hawaiian monk seals eliminating duplicative research. The worming trial may share animals with foraging research to reduce human activity and the number of animals be utilized in monk seal research and conservation actions. However, we have several partners aiding us in the analysis of our samples and data. These include: Bishop Museum, Moss Landing Marine Lab, University of Hawaii Manoa and Hilo, UH Hawaii Institute of Marine Biology, Southwest Fisheries Science Center, Scripps Institute of Oceanography and Dalhousie University, Canada.

Data collected during this study will also be provided to the Monument to aid with their management objectives.

**12a. List all specialized gear and materials to be used in this activity:**

none

**12b. List all Hazardous Materials you propose to take to and use within the Monument:**

Separate attachment will accompany this application.

**13. Describe any fixed installations and instrumentation proposed to be set in the Monument:**

No permanent fixed installations will be set in the monument for this work.

**14. Provide a time line for sample analysis, data analysis, write-up and publication of information:**

Tissue samples will be analyzed at different times. Feces, blubber and other tissues used for diet analysis will be processed and logged within one month of return to Honolulu. They will then be distributed to the appropriate lab for analysis. Other samples should be analyzed within 6 months of collection depending on the workload of partner and contract laboratories. An important point to emphasize is that we do have partners in place to analyze samples and interpret resulting data. Analysis results will not occur until after the second year of the deworming trial has been completed (by May 31, 2011). Publication of results is expected in the year following the analysis.

**15. List all Applicants' publications directly related to the proposed project:**

With knowledge of the penalties for false or incomplete statements, as provided by 18 U.S.C. 1001, and for perjury, as provided by 18 U.S.C. 1621, I hereby certify to the best of my abilities under penalty of perjury of that the information I have provided on this application form is true and correct. I agree that the Co-Trustees may post this application in its entirety on the Internet. I understand that the Co-Trustees will consider deleting all information that I have identified as "confidential" prior to posting the application.

---

Signature

Date

**SEND ONE SIGNED APPLICATION VIA MAIL TO THE MONUMENT OFFICE  
BELOW:**

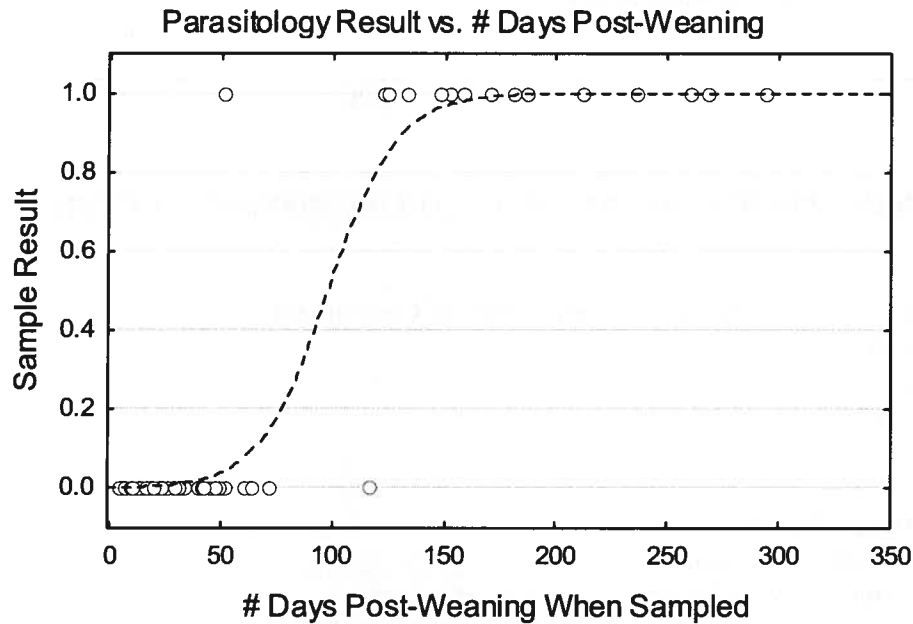
Papahānaumokuākea Marine National Monument Permit Coordinator  
6600 Kalaniana'ole Hwy. # 300  
Honolulu, HI 96825  
FAX: (808) 397-2662

**DID YOU INCLUDE THESE?**

- ☐ Applicant CV/Resume/Biography
- ☐ Intended field Principal Investigator CV/Resume/Biography
- ☐ Electronic and Hard Copy of Application with Signature
- ☐ Statement of information you wish to be kept confidential
- ☐ Material Safety Data Sheets for Hazardous Materials

## Appendix A: Infection chronology for weaned monk seal pups.

Figure 1: Probability of parasite infection in weaned monk seal pups versus number of days since weaning. Plot shows a logistic regression for probability of positive result for one or more of three parasite infection assays (*Diphyllbothrium spp.*, *Contracaecum*, *Acanthocephalans*) versus number of days since weaning. All but one pup weaned more than 100 days earlier tested positive. Results are from fecal loops collected during health screening in 1998-2000 (n=54 seals).



## APPENDICES 1-5

### Appendix 1: Hawaiian Monk Seal Captive Care Protocol for Kewalo Research Facility, 2007-2008

#### I. QUARANTINE

##### A. QUARANTINE DEFINITION AND OBJECTIVES

1. Quarantine refers to “any isolation or restriction on travel or passage imposed to keep contagious diseases, insect pests, etc. from spreading (ref).”
2. Hawaiian monk seals currently held in captive care must be maintained under strict quarantine at all times to:
  - a. Minimize transmission of disease from outside sources- i.e. human contact
  - b. Minimize transmission of disease from captive care seals to susceptible animals, including wild seals, humans, etc.
  - c. Minimize transmission of disease among the three holding tanks at Kewalo Research Facility (KRF)
3. All personnel involved in the feeding, handling, and care of these seals must be properly trained in quarantine procedures by an experienced staff. (Quarantine procedures should always be posted in the food preparation area.)

##### B. NMFS QUARANTINE POLICY

###### Quarantine from Outside Sources

1. All equipment used in the quarantine facility, including feeding, handling, and medical supplies MUST be:
  - a. Labeled “MONK SEAL QUARANTINE”
  - b. Used exclusively for quarantined seals
  - c. Kept monk seal equipment separate from that used for other animals
  - d. Properly sanitized before and after entering the quarantine enclosures
2. **NO VISITORS** are allowed in monk seal quarantine area unless previous approval is granted by the on-site supervisor. Non-authorized personnel may be able to view the seals from an approved viewing platform outside the quarantine area.
3. Avoid direct contact with domestic or other captive or wild animals before and after entering Hawaiian monk seal quarantine enclosure. Shower and change clothes before and/ or after going to another animal care facility if entering the seal enclosures on the same day.
4. No street shoes are to be worn into the enclosures, including the walkway leading to tanks 1 and 2. Wear rubber boots/shoes designated for “monk seal quarantine” use in the enclosures at all times. Minimize wearing quarantined boots/shoes around KRF premises.
5. Dip soles of boots/shoes in dilute Nolvasan footbath upon entering AND leaving all the enclosures.
6. Immediately upon entering the enclosure to tanks 1 and 2, wash hands with antibacterial soap. Also wash hands before and after fish preparation, feeding, or handling seals. Always wash hands immediately after leaving a separate tank enclosure. Before and after entering tank 3, wash hands at sink next to the enclosure.
7. Any person that will potentially come in direct contact with quarantined seals for any procedure must wear sanitary protective clothing (i.e. coveralls, handling gloves, shoes) designated for quarantine monk seal use only. This clothing should be kept clean and in a designated area away from potential sources of contamination.
8. Protective clothing worn during procedures should be immediately washed in the washer with soap and dilute bleach solution following handling events.
9. Any new equipment or tools brought into the quarantine area must first be sanitized with a dilute bleach or Nolvasan solution.

### **Quarantine Between the Three Holding Tanks at KRF**

1. Separate equipment will be used to care for seals in each of the three tanks. This includes cleaning and feeding supplies (brooms, hoses, buckets, etc.) and handling gear (coveralls, booties, gloves). Keep this equipment separate.
2. A dilute Nolvasan footbath will be placed at the door of each tank to be stepped before and after leaving the enclosure. A freshwater bath will be placed as a final rinse before entering the pen.
3. Personnel must change protective clothing when caring for seals housed in tanks 1 and 2. However, personnel caring for isolated seals in tank 3 are prohibited from entering the "healthy" seal area (the stairs, walkway, fish house, and tanks 1 and 2).
4. Seals housed in separate enclosures will not be mixed unless deemed necessary by the veterinary staff.
5. A seal require isolation, follow the Potential Disease Outbreak Protocol.

## **II. OBSERVATIONS AND CONDUCT AROUND SEALS**

### **A. OBSERVATIONS OF THE SEALS**

1. In the morning and prior to each feed, conduct a thorough inspection of the seals and pens before proceeding with further activity. Following each feed or handling event, monitor the seals' behavior closely. Perform a final inspection before leaving for the day.
2. Throughout the day monitor and record the behavior of each seal. Observe the condition and activity level of the seals and presence of feces, urine, spew, and harmful debris in or around pens. When possible, note ID of seal that produced scat, spew, etc. Note the color, consistency, and amount of scat, urine, and spew.
3. Note anything unusual in a seal's normal appearance (eyes, nasal discharge, bite wounds, etc.) and behavior (lethargic, unresponsive, etc.). Notify attending veterinarian and animal care manager immediately of any abnormal changes in a seal's health.
4. Succinctly record any observations on the "Observation" form in each seal's chart, including the time and observer's initials. Frequently used acronyms: BAR = bright, alert, and responsive; QAR = quiet, alert, and responsive.

### **B. CONDUCT AROUND THE SEALS AT ALL TIMES**

Every possible effort should be made to minimize the habituation of the seals by reducing human-seal interactions.

1. When in enclosures, **DO NOT MAKE PHYSICAL CONTACT WITH SEALS** unless necessary for procedures requiring handling. Minimize going into the enclosure and the amount of time you spend in the enclosure as much as possible.
2. If seals are resting or sleeping, do not make loud noises or startling gestures, and move slowly when in close proximity to them to minimize stress.
3. Minimize talking when working with or near the seals and the enclosure.
4. Whenever possible, observers should remain as inconspicuous and unobtrusive as possible to observe seals' normal behaviors in captivity and minimize their stress in captivity.
5. Each person entering an enclosure with the seal should be carrying a herding board, which should be within arms-reach at all times.
6. Outside of feeding sessions seals may display undesirable behaviors which include: a) approaching too closely or too rapidly; b) mouthing hoses, brooms, or boots; and c) stereotypic behaviors which include repetitive splashing or slapping at the walls of the enclosure. If seals approach too closely or too rapidly use a herding board to keep the seal away. The mouthing of brooms, hoses, and boots should be discouraged by preventing opportunities for seals to bite at these objects in the first place. Stereotypic behaviors are a sign of boredom and may be reduced by providing seals with their approved environmental enrichment devices (EEDs).

### **III. CLEANING THE QUARANTINE AREA**

#### **A. DISHES**

1. Wash all dishes used for feeding and handling with dish soap and water. Rinse thoroughly.
2. Soak all metal and rubber equipment (bolus syringes, knives, tongs, etc.) in dilute Nolvasan for at least 10 minutes.
3. Soak all plastic equipment (cutting boards, buckets, cooler, etc.) in dilute bleach for at least 10 minutes.
4. Rinse all dishes thoroughly to remove the dilute bleach or Nolvasan.
5. Allow all dishes to air-dry.
6. Stomach tubes should be washed with soap and water, rinsed thoroughly, and then boiled for 10 minutes. Be sure to scrub the inside of the feeding tubes. Keep sanitized stomach tubes wrapped in a clean towel.
7. Bolus Syringe Care: after the syringes have been washed and dried as described above, lubricate the O-ring with mineral oil and put the syringes back together for safe storage. Be careful when handling the syringes as they are fragile and can crack easily.

#### **B. DAILY CLEANING AND MAINTENANCE**

##### **Seal Enclosure Cleaning**

Do not allow seals to mouth or bite brooms or fresh water hoses. If the hose enters the pool remove it immediately. Never allow the broom, hose, or any equipment to remain unattended in a seal enclosure. Return all equipment to its storage area after use (i.e. coil and hang hose). Always keep the enclosure doors securely bolted because the seals are very adept at exiting the enclosures through a door left ajar. When cleaning, take the opportunity to inspect urine for color and feces for consistency and parasites. Always record feces and urine in the observations form in the seal's chart and make special note of any unusual findings.

1. After the morning feed, the entire pen enclosure should be checked for any scat, urine, fish parts, and wind-blown debris. If necessary, use a broom and fresh water hose to clean the seal enclosure. Thoroughly rinse all fish scales, blood, and debris from the decks, walls, and ledge of the enclosure and walkway with the fresh water hose after each feed. Special care should be taken to clean scales from doors, door handles, and bolts.
2. Before leaving in the evening, the deck and pool walls and floor should be hosed down and any spattered blood, scales, scat, or other debris should be scrubbed away.

##### **Miscellaneous Cleaning**

1. Rinse off the walkway and stairs leading to the seal enclosure at least once a day. Scrub the walkway with broom and water as needed.
2. Refill footbaths 1-2 times a day or as needed with dilute Nolvasan (usually once first thing in the morning is fine). Add 3 oz Nolvasan to 1 gallon water. Be sure to have a final water rinse before the pen entrance. Old dilute Nolvasan should be poured onto the pavement next to and at the base of the stairs (don't dump it down the stairs).

##### **Fish house Cleaning**

1. Freezers and refrigerators must remain clean and neat at all times. All feeders are responsible for maintaining freezer cleanliness on a daily basis. Keep freezers free of ice buildup as much as possible.
2. Wipe down all counter and table surfaces after each feeding. Be especially mindfully of cleaning any fish scales and spattered blood from the all surfaces after each feeding.

3. Mop the fish house floor with a dilute bleach solution (1 part bleach to 30 parts water) after the morning feeding.
4. Empty the garbage and take it to the outside dumpster at the end of each day (or every other day).
5. All damaged or unused fish and fish parts including the scales should be put in the "Fish Waste" bag in the chest freezer and should NOT be thrown away in the regular trash. The "Fish Waste" bag should be taken to the facility dumpster every Wednesday before 0800 for immediate pick-up.

#### **Coverall Cleaning**

1. Wash all coveralls, kneepads, gloves, and booties following each use with dilute bleach and laundry detergent in the washing machine at the end of each day. Do not set quarantine items down outside the quarantine area.
2. Dry all items in the dryer except the booties with rubber soles. The booties should be air-dried on the floor in the fish house.
3. In between handling events on the same day, hang the coveralls in the sun to dry.
4. Store clean, dry coveralls, etc. in the box in the fish house labeled "Clean Handling Equipment" (in the cabinet).

### **C. WEEKLY CLEANING**

#### **Cleaning Seal Enclosure**

The monk seal tanks and enclosures are drained and cleaned once a week. A minimum of 3 people should conduct the weekly cleaning. Pool cleaning occurs every Friday following the 0800 feed. A dilute bleach solution should be used (the black sprayers holds 2 gallons so add about 1 cup of concentrated bleach). When using bleach solutions always direct the rinse water away from seals because the bleach solution is a skin and eye irritant. Use the large, soft-bristled brushes for all cleaning.

1. Empty all Nolvasan footbaths except for one at the base of the stairs. Once the footbaths are emptied, minimize leaving and reentering the quarantine area as much as possible.
2. Spray the walkway with the dilute bleach mixture. Be sure to spray the walls, ledge, and doors. Direct the bleach spray away from the seal enclosure. Scrub the walkway deck and ledge. Let the bleach stand for 10 minutes.
3. Hose off the dilute bleach thoroughly – perform at least 2 washes of all surfaces.
4. When you are convinced that all the bleach has been rinsed away, move the seals into the holding area. Use a net to scoop the seals out of the water and herding boards to direct them into the holding area. Be sure to keep the seals cool with running water while cleaning the enclosure and monitor the seal's affect and behavior regularly.
5. Once the seals are secure in the holding area, begin draining the pool and start bleaching the deck. Spray the entire deck and up to the fiberglass line on the walls. Be careful to avoid sending bleach (from the sprayer or wind) into the seals' holding area. Spray the cages, any enrichment tools, and drain covers. Scrub the deck floor and allow the bleach to stand for 10 minutes.
6. As the pool continues to drain, thoroughly rinse the bleach from the deck area. Perform at least 2 rinses of all surfaces.
7. When the pool is drained, scrub the walls and floor of the pool to remove all scales and spattered blood. Using the hand-held brushes works well for scrubbing the walls.
8. Spray the pool walls and floor with dilute bleach and scrub all surfaces again. Let the bleach stand for 10 minutes.
9. Rinse the bleach from the pool walls (at least twice) while simultaneously turning on the water inflow. Leave the bottom drain open, with the drain cover in place, for several minutes to thoroughly rinse all the bleach down the drain. When you feel confident that all the bleach has been washed away, close the bottom drain, and begin filling the pool.



10. Flush the deck for several minutes to remove any bleach remnants. Remove all cleaning equipment then bring the water level with the deck and re-introduce the seals to the enclosure.
11. After all the tanks and walkways have been cleaned, thoroughly rinse the bleach solution from the brooms and all cleaning equipment. Separate cleaning supplies are used in each of the tanks. Replace the Nolvasan solution (3 oz/1 gal) in all footbaths.
12. Record the seals' behavior, the duration spent in the holding area, and any other relevant information from the cleaning event (scat, spew, urine, etc.) on the observations form in each seal's chart.

#### IV. WATER SAMPLING SEAL TANK

Sampling should occur on the same day (Tuesday) and time (1000-1100) each week at least a couple of days after the weekly enclosure cleaning. We collect one sample from the pool and one from the inflow in addition to a temperature control sample collected from the pool. These samples are submitted to Hawaii Food & Water Testing Lab (HF&WTL) for fecal coliform testing.

1. Try to be as sterile as possible: wear gloves, do not open lid to bottle until immediately before collection, do not contaminate inside of lid or bottle, don't set the lid down, etc.
2. Collect the inflow sample by removing the lid and holding the bottle under the water inflow to fill it. Decant any excess water being careful not to touch the lip of the bottle or the lid.
3. Sample the pool (pool and temp control sample) 180° from the water inlet. With the lid still in place, submerge the bottle about 1 foot deep. Unscrew the lid underwater with the bottle positioned counter-current to fill the bottle. Replace the lid underwater. Remove the bottle from the water and decant the excess water being careful not to contaminate the bottle or lid.
4. Immediately place the samples in the small red cooler with blue ice (provided by HF&WTL) for transport to the lab. If transport is not immediate, place the samples in the refrigerator (sampling fridge, not fish storage fridge). Store sample bottles in the cooler and ice pack in freezer until next sampling.
5. Complete all the necessary paperwork and be sure to label each bottle (pool, inflow, temp control).
6. Results submitted on Tuesday are usually faxed to us, c/o Robert Dollar, on Thursday or Friday. These counts should not exceed 1000 MF/100ml. If fecal coliform counts exceed 1000 MF/100ml, results are reported to Robert Dollar by phone; sampling must be repeated within 24 hours. Promptly notify the veterinary staff if counts are above 1000 MF/100ml.
7. Enter the date, time, coliform count, and any pertinent comments in the HMS Water Testing spreadsheet.



#### V. SEAL ILLNESS/EMERGENCY CARE

1. In case of an emergency or suspected illness, refer to the phone list and call the attending veterinarian or veterinary technician immediately to relate symptoms or circumstances of emergency or illness. Follow the emergency chain-of-command protocol.
2. A veterinarian or trained veterinary staff will perform any needed blood sampling.

A crash kit and emergency drugs are kept in the fish kitchen. All other medical supplies for blood sampling, fluid and antibiotic administration, monk seal medications, and additional medical supplies are kept at

## Appendix 2: Hawaiian Monk Seal Feeding Protocol

- Begin tube feeding the seal at the next scheduled feeding time after admit or as determined by the veterinary staff.
- Weaners: Feed the seals three times per day with 5-6 hours between each feed.
- Yearlings: Tubings may be reduced to twice a day to reduce stress, if determined by the veterinarian.
- The seals will be tube feed until they begin eating dead herring on their own.
- All seals will be re-hydrated with electrolyte fluids for the first 24 hours. After that time, a diluted fish mash slurry will be introduced followed by full-strength fish mash. The volume to be tubed is determined by admit weight as described below.
- Begin offering yearlings fish prior to tube feedings 36 hours after admit.
- Begin offering weaners fish prior to tube feedings 72 hours after admit.
- Initially offer 0.25 kg of fish at each scheduled feed before attempting to tube feed. If the seal does not eat the entire amount of fish offered, then tube feed according to the weight specifications.
- Once the seal has consistently eaten 0.25 kg of fish at 3 feeds, the seal is now considered a free feeder. Begin increasing the feed amount by 0.25 kg every day at each feed until a daily intake of about 6-7% the seal's body weight is reached.
- After the first 72 hours, if a seal is not eating 2 days later, begin increasing the tubing volume by 50 mL every day at each feed. The maximum tubing volume will be determined by the seal's weight and response to the increased volume at each tubing.

### Feeding Schedule for First 72 Hours

Feed #	Admit Weight				Notes
	< 30 kg	30-39 kg	40-50 kg	> 50 kg	
1	300 mL EL	500 mL EL	750 mL EL	1000 mL EL	
2	300 mL EL	500 mL EL	750 mL EL	1000 mL EL	
3	300 mL EL	500 mL EL	750 mL EL	1000 mL EL	
4	150 mL EL 150 mL FM	250 mL EL 250 mL FM	375 mL EL 375 mL FM	500 mL EL 500 mL FM	
5	150 mL EL 150 mL FM	250 mL EL 250 mL FM	375 mL EL 375 mL FM	500 mL EL 500 mL FM	
6	150 mL EL 150 mL FM	250 mL EL 250 mL FM	375 mL EL 375 mL FM	500 mL EL 500 mL FM	Yearlings: begin offering fish, tube if not eating
7	300 mL FM	500 mL FM	750 mL FM	1000 mL FM	
8	300 mL FM	500 mL FM	750 mL FM	1000 mL FM	
9	300 mL FM	500 mL FM	750 mL FM	1000 mL FM	Weaners: begin offering fish, tube if not eating

EL = Electrolytes  
 FM = Fish Mash

### ELECTROLYTE (EL) SOLUTION

A soluble powder electrolyte supplement (BounceBack®, Manna Pro, St. Louis, Missouri) will be used to help the seals retain essential body fluids.

1. Mix 1 scoop of BounceBack with 2 quarts of filtered water.
2. Stir until the powder is completely dissolved.
3. When finished, pour the solution into a clean container and label it with “electrolytes” and the date, time and your initials.
4. Store in the refrigerator for up to 24 hours.

### **FISH MASH (FM) PREPARATION**

Fish mash is equal parts fish and filtered water blended together using a commercial-strength blender. For example, you would weigh out 1 kg of chopped fish and add 1000mL of water. About 2 batches of fish mash can fit in the blender at a time. More concentrated fish mash can be made or additional sources of fat (salmon oil) may be added if necessary as directed by veterinary staff.

1. Cut off the heads, fins, and tails from slightly defrosted fish. Chop the fish into small pieces.
2. Weigh out 1 kg of fish pieces.
3. To blend, add 1000mL of water and about half the amount of chopped fish pieces. Make sure the fish is not too frozen as this will wear out the motor on the blender. Do not run the blender or more than one minute at a time.
4. Blend on low for 10 seconds.
5. Add the rest of the fish pieces.
6. Blend on low for 10 seconds then on medium for 30-45 seconds.
7. Stir the fish mash with spatula to make sure there are no large chunks. If there are any chunks that might get stuck in the stomach tubes, blend again.
8. When finished, pour the fish mash into a clean container and label it with the type of formula, date, time and your initials.
9. Store in the refrigerator for up to 24 hours.

### **FISH PREPARATION**

IQF herring is stored in the chest freezer in the Fish House and thawed on an as-needed basis. All frozen fish must be well sealed to prevent freezer burn and drying. Always re-tie the plastic bags of herring after each use. Any fish that are broken or “damaged” should not be used.

1. Review feed amounts in each seals’ chart before preparing fish. This information is also posted on the whiteboard in the fish house.  
Note: Check the orders form for instruction on fish prep for each seal. For example, you may need to cut off the head and pectoral fins (on a diagonal), the anal and dorsal fins, and tail.
2. Wash hands thoroughly with antibacterial disinfectant soap before handling buckets and fish. Always wear gloves when preparing feeds.
3. Thaw the fish in running water. Make sure the drain covers are in place so scales don’t clog up the pipes. While thawing, inspect the fish again for bruises, cuts, or other abnormalities not detectable when frozen. Discard any damaged or broken fish.
4. Throw accumulating scales from the drain covers and all unusable fish parts in the plastic bag in the chest freezer labeled “Fish Waste.” These will be put in the facility dumpster just prior to trash pick-up early Wednesday morning.
5. Weigh the fish using the digital scale and each seal’s feeding pail. Turn the scale “ON” and allow it to zero. Put the pail on the scale and let it equilibrate, then push the “TARE” button. Tare the scale before each seal’s feed. Record the feed weights in the feeding log in each seal’s chart.
6. If supplementing the herring feed with a multivitamin (Pinnivite = PV) insert the vitamin tablet into the gill slit or body cavity of one of the fish.

7. If not feeding immediately, keep the fish cool in the refrigerator or on blue ice in a cooler until ready to feed. Keep feed amounts for each seal separate.
8. Fish can be defrosted in the refrigerator prior to use but once thawed, the fish are only good for a maximum of 24 hours in the refrigerator. Thawed fish should be labeled with the date and time that the fish was defrosted.

### **TUBE FEEDING**

Various methods can be used to tube feed a seal. Use the method that the feeder and restrainers are most comfortable with, decided on before entering the seal enclosure. The minimal number of people should be used to restrain the seal.

1. Wash hands with disinfectant soap immediately prior to and after feeding.
2. Prepare the electrolyte solution, fish mash, or other formula (check in the refrigerator as there may be some already made within the last 24 hours).
3. Measure out the specified amount according to the seal's chart (measure out a little extra to account for fish mash lost before the tubing). This information is also posted on the whiteboard in the fish house. Return any unused electrolytes/fish mash to the refrigerator.
4. Warm up the electrolytes/fish mash for 30 seconds in the microwave. Thoroughly stir the fish mash to distribute the heat. A second round of warming may be required to remove the chill (be careful not overheat).
5. Ground up the vitamin and any other supplements or medications with a mortar and pestle. Pour this into the electrolytes/fish mash and stir thoroughly.
6. Draw up the electrolytes/fish mash into 1-2 500 mL bolus syringes.
7. Put a feeding tube, the 1-2 bolus syringes (keep upright), and any remaining electrolytes/ fish mash (in a beaker) for the feed into a bucket to be taken into the seal enclosure.
8. Anyone entering the enclosure to handle the seals should be wearing gloves, coveralls, and booties and step in Nolvasan footbath and freshwater rinse before entering pen. Use a separate set of coveralls when working in each seal enclosure. Have a clear plan of action before entering the pen.
9. Keep the seal cool during and post-handling. Hose off post-handling if necessary. Restrain and feed in the shade if possible.
10. Enter the enclosure slowly and minimize startling the seals.
11. Once the restrainer(s) have the seal under control (use a towel and herding boards), the feeder inserts the tube into the mouth of the seal. The head restrainer may need to "pop the jaw" by inserting his/her thumbs or forefinger between the jaw behind the molars (outside the mouth).
12. Slowly insert the tube into the stomach, allowing the seal to breathe normally.
13. Ensure that the tube is in the stomach before administering the electrolytes/fish mash by listening for stomach gurgling. Blowing gently on the end of the tube may be required to hear these sounds.
14. Gently and steadily plunge the electrolytes/fish mash into the stomach. Crimp the tube when removing a bolus syringe from the stomach tube so as not to create a vacuum. The stomach tubes hold about 50 mL so after administering all the electrolytes/fish mash, plunge about 50 mL of air through the tube.
15. Slowly remove the stomach tube.
16. After the tube feeding, release the seal and exit the enclosure immediately. Remove all feeding equipment. From outside the pen, watch closely for regurgitation, scat, and post-handling behavior.
17. Record observations associated with the feeding and handling in each seal's chart (in the Observation form) and record the amount feed in the Dietary/Medical Record form.

18. Perform all post-feeding cleaning duties including: washes all dishes, cleaning all surfaces in the fish house, and mopping the fish house floor (morning feed only).

### **OFFERS AND FISH SCHOOL**

There are a variety of techniques that can be used to encourage the seals to begin free feeding as quickly as possible, which are outlined in Guidelines for Offers and Fish School.

### **FREE FEEDING**

1. Wash hands with disinfectant soap immediately prior to and after feeding.
2. From the walkway between tanks 1 and 2, throw fish to each seal at a rapid pace to minimize begging behavior. Throw a handful of fish at one time to always have at least a couple of fish waiting in the water for the seal to eat so that the seals are looking in the pool for fish. Don't throw a fish when the seals are looking or vocalizing at you. Try to keep the seals on opposite sides of the pool as much as possible. It is preferable that each seal eat only the fish from their bucket but if they steal each other's fish, swap fish of similar size from the other's bucket to make sure each eats their share.
3. Give the vitamin fish after observing that the fish are being eaten (second or third fish). Closely observe if vitamins are ingested or sink to bottom of the pool. If a seal does not receive vitamins, record this in the seal's chart (on the dietary/medical record and observations forms) and leave a note that they should be given at the following feed. Notify the veterinary staff ASAP if medications are not received.
4. Any uneaten fish should be disposed of in the "Fish Waste" bucket in the chest freezer. Scooping the pool for uneaten fish pieces is often unnecessary but if large chunks of fish are not consumed, scoop these from the pool and weigh this amount (subtract from the amount fed when recording in dietary record form). Be sure to clean, bleach, and thoroughly rinse the net after each use.
5. Record observations associated with the feeding in each seal's chart, including any adverse behavior (on the Observations form). Record how much each seal actually ate, estimating weight of additional fish ingested or fish lost when necessary, in the Dietary/Medical Record form. Frequently used acronyms: AWIP = ate well in pool.
6. Perform all post-feeding cleaning duties including: hosing off splattered blood and scales, washing all dishes, cleaning all surfaces in the fish house, and mopping the fish house floor (morning feed only).

### **Appendix 3: Hawaiian Monk Seal Medical Care of the Seals**

The medical treatment regime for a sick or injured seal will be determined on a case-by-case basis by the veterinary staff. Detailed observations by caregivers will greatly facilitate medical decision-making. The following factors will be considered to help determine the appropriate treatment regime for a seal:

- Overall attitude – activity level and behavior (lethargic, depressed, quiet, lying curled up, hunched posture, logging in the water, active, alert, interacting with other seals, swimming, hauled out, etc.)
- Presence of vomit, urine, or feces (amount, color, and consistency)
- Appetite and feeding behavior (loss of appetite, no interest in fish, loss of interest abrupt or gradual)
- Experience with seals with similar clinical signs (with Hawaiian monk seals and other pinnipeds handled at TMMC)

#### **Daily Medical/Physical Examination**

A complete medical/physical examination will be performed daily using the Physical/Medical Exam Form to record data. Seals WILL NOT be restrained for this physical exam; it will be strictly a visual exam. If any abnormalities are found, notify the veterinary staff immediately. A more thorough description of any abnormalities should be documented in the Medical Progress Report by the veterinary staff. Also complete the Wound Sheet or Eye Exam Form if applicable. Photographs of any physical abnormalities will also be taken. The course of treatment and sample collection of all abnormalities will be determined by a committee of veterinarians and experienced monk seal caretakers.

#### **Frequently Encountered Medical Problems and Treatments**

##### **Malnutrition**

For all underweight seals, the objective is to maintain a positive energy balance and good hydration status. A sign of good hydration is tearing eyes (wet circles around the eyes). Weekly weights will be used to assess if the caloric needs of each seal are being met. Severely underweight seals may be too weak to fight infections or other illness so all efforts will be made to eliminate any factors that might compromise the health of the seal, most of which are standard operating procedures: limiting exposure to the elements (both heat and cold), reducing the stress on the seal, maintaining quarantine.

- Follow the appropriate feeding protocol.
- It may be necessary to use a formula with a higher caloric value (see Feeding Protocol) or increase the number of feedings per day.
- Subcutaneous fluids with dextrose may be administered to provide additional calories.
- If a seal is still not gaining weight although eating an adequate number of calories, check the blood work to investigate potential underlying causes.
- Anthelmintics, which may improve absorption, will be administered to reduce the load of gastrointestinal parasites when the seal is hydrated.

##### **Stress**

During past captive care efforts with Hawaiian monk seals, some seals, especially yearlings and other older seals, have exhibited signs of acute stress. Clinical signs of stress include stereotypic behavior, which is classified as any repetitive motor behavior that does not have an obvious purpose or function. Examples of stereotypic behaviors include pacing and swimming in circles. Stress may inhibit the seal's

ability to fight underlying infections or illnesses; every effort should be made to reduce the stress of the seal and support the seal nutritionally.

- Maintain a positive energy balance and good hydration status as outlined above.
- Maintain normal body temperature (protect from the elements)
- In extreme cases, the activity level of the seal may need to be limited by minimizing or restricting access to the swim area. This approach will be used very cautiously so that it does not further stress the seal.
- A change in the group size or structure may be tested for its effects on seal stress behavior.
- Medication to reduce the stress level of the seal may be administered as prescribed by the attending veterinarian. Such medications include diazepam and haloperidol.

### **Periods of Inappetence**

Once free feeding, a seal may stop eating at any time. Satiation, warm weather, stress, or ill health may cause inappetence. If satiation is the cause, the seal should quickly resume free feeding after cutting its dietary intake for a brief period (each seal has a different satiation point; the feed orders will be adjusted for each seal so as not to exceed this limit).

- If a seal does not eat at 1 feeding session, monitor the behavior and overall affect of the seal. At the next feed, offer the normal feed amount. Monitor the feeding behavior closely during this feeding session.
- If the seal is still not eating after 2-3 feeds, complete a medical examination, draw blood and tube electrolytes at the same time (tube volume according to weight specifications). It is important to re-hydrate the seal with several electrolyte tubings before returning to offering fish.
- Subcutaneous fluids may also be administered as determined by the veterinary staff to help the seal re-hydrate more quickly.
- A rectal swab (Cary Blair media) or fecal sample may be collected if a gastrointestinal bacterial infection is suspected as the cause of the inappetence.
- Proceed with offers before tube feedings using an abbreviated 72-hour feeding schedule as determined by the veterinary staff (may increase the tubing/offer amount more rapidly).

### **Multiple vomiting episodes**

Vomiting episodes may accompany periods of inappetence. If spew is discovered in the pen, make every effort to identify the seal that produced the spew. Monitor all the seals in the pen at subsequent feedings for any signs of inappetence to help identify this seal.

- For an isolated instance of vomiting, monitor the appetite and behavior of the seal. Carefully check for additional spew.
- If multiple vomiting episodes occur, stop feeding the seal for 12 hours. Administer 1 shot of DualPen (1 ml/10kg IM) during the first 12 hours of not eating to treat for possible Clostridium infection. If signs do not resolve, draw blood and begin a 7-day treatment of DualPen (1 ml/10kg IM every other day).
- Support the seal nutritionally with tube feedings until it is eating again.
- A rectal swab (in Cary Blair media) or fecal sample may be collected if a gastrointestinal bacterial infection is suspected as the cause of the vomiting.
- Other medications and subcutaneous fluids may be administered as determined by the veterinary staff (such as Pepto-Bismol, cimetidine, metaclopramide).

### **Abscesses**

Seals may be admitted with abscesses or may develop them while in our care, usually from bites by other seals. Treatment depends on the site and severity of the abscess.



- Lance the abscess with a sterile scalpel blade, swab the inside of the abscess with a culturette and place in Amies transport media, and flush with dilute chlorohexadine or hydrogen peroxide every other day until no more pus is visible at the drainage site.
- For small abscesses (quarter-sized or smaller), antibiotics or other medications will not be administered if the seal is acting healthy.
- For large abscesses, treat with antibiotics, typically amoxicillin, according to the dosages on the Drug Formulary.
- If the abscess recurs, check the culture and sensitivity results of the initial swab for choice of antibiotic treatment.

#### **Eye lesions**

The eyes will be closely monitored during the daily medical/physical examination.

- If any signs of ocular disease develop, start daily ocular exams using the Eye Exam Form to record data.
- Four swabs of the cornea and conjunctiva will be collected. One swab will be used for bacterial culture, one for herpes PCR, the others banked at -70°C.
- Topical antiviral and antibiotic treatment will be started immediately according to the Eye Disease Protocol
- Isolate the seal(s) with ocular diseases and maintain strict quarantine.
- Commence rounds with a veterinary team to monitor treatment and progress. Frequency of rounds will vary, but will be at least once a week.

#### **Appendix 4: Hawaiian Monk Seal Sampling Protocol**

The Sampling and Morphometric Checklist Form must be completed for all admit, re-check, and release exams.

##### *Admit Exam*

All seals collected for inclusion in captive care will undergo an admit exam 1-3 days after being collected. During this exam, the seal will be physically restrained and a blood sample, fecal sample, 2 swabs of each orifice (eye, nasal, oral, rectal, and genital), standard length, axillary girth, and body weight will be collected.

##### *Re-check Exams*

- If a seal is clinically healthy and has normal blood work (values within normal ranges for Hawaiian monk seals), blood will be drawn and re-checked every 4 weeks (to make sure blood work is still normal and to bank a series of control samples for each seal).
- If a seal has abnormal blood work, blood will be draw and re-checked every 1-2 weeks until normal levels are attained.
- If a seal is showing signs of ill-health, obtain a blood sample as soon as possible and re-check every 1-2 weeks until normal levels are attained.
- If any abnormalities are present, additional blood sampling may be requested by the veterinarians. Other samples (swabs, biopsies, etc.) may also be requested by the veterinarians.

##### *Release Exam*

All seals included in captive care will undergo a release exam 4-7 days prior to release. During this exam, the seal will be physically restrained and a blood sample, fecal sample, 2 swabs in each orifice (eye, nasal, oral, rectal, and genital), blubber biopsy, standard length, axillary girth, and body weight will be collected.

##### *Blood sampling*

- Collect up to 10 mL of whole blood from the extradural venous sinus during each exam. A larger volume of blood may be collected once the seal is hydrated, at the release exam, or as determined necessary by the veterinary staff for diagnostic purposes.
- Collect 1 EDTA lavender top tube (LTT) for CBC. Fill the LTT half way and immediately but gently invert 5-6 times to mix additive with blood (ensure the blood is not clotted). After CBC testing is complete, spin the tube in a centrifuge for 10 minutes and pipet off the plasma and bank (label PS LT). Also pipet off and bank the buffy coat (label BU LT).
- Collect at least 2 serum separator tubes (SST) for serum chemistry and serum banking. Do not invert SST's. Let SST tubes sit at room temperature for at least 15-30 minutes (but not more than 1 hour); then spin tubes in centrifuge for 10 minutes. Pour off the serum into 1 mL aliquots (label SE).
- Collect 1 heparin green top tube (GTT) for serum chemistry testing using an I-Stat (for immediate estimates of blood values). Fill the GTT half way and immediately but gently invert 5-6 times to mix additive with the blood (ensure the blood is not clotted). After I-Stat testing is complete, invert the GTT 5-6 times to re-mix and pipet off 0.1 mL blood into a cryovial to be banked for biotoxin testing. Also apply a drop to each of the eight circles on the biotoxin card (see NMFS Hawaiian Monk Seal Sampling Protocol). Spin the tube in a centrifuge for 10 minutes and pipet off the plasma and bank (label PS GT). Also pipet off and bank the buffy coat (label BU GT).

- All banked samples should also be labeled with the seal's permanent ID, date collected, and specimen #.
- Complete Blood Count (CBC) testing: white blood cell count, white blood cell differentials, red cell count, hemoglobin, hematocrit, mean corpuscular volume, mean corpuscular hemoglobin, mean corpuscular hemoglobin concentration, platelet count
- Serum chemistry testing: sodium, potassium, chloride, blood urea nitrogen, creatinine, glucose, calcium, phosphorus, uric acid, total protein, albumin, globulin, osmolarity, total/direct/indirect bilirubin, gamma glutamyl transpeptidase, lactate dehydrogenase, aspartate aminotransferase (serum glutamic-oxaloacetic transaminase), alanine aminotransferase (serum glutamic-pyruvic transaminase), alkaline phosphatase, iron, cholesterol, triglycerides, creatine phosphokinase, amylase, lipase, fibrinogen

#### *Fecal examination*

- One fecal sample will be examined for presence of parasite ova by fecal float. Samples can be held at 4°C if examined within one week of collection, otherwise they should be preserved by addition of equal volume of formalin to the fecal container.
- Aerobic culture of feces will be done on admit, release and any diarrhea samples as directed by the veterinarian. Swabs for fecal culture should be placed in Cary Blair transport media and stored at 4°C if cultured within one week of collection, otherwise frozen at -70°C until culture is possible.

#### *Swabs*

- Swabs of each orifice should be stored in dry cryovials at -70 C

## **Appendix 5: Contingency Plans for Hawaiian Monk Seals Held at Kewalo Research Facility**

As with an emergency situation, the safety of the personnel will be the first priority followed by the safety and health of the seals. Animal care and quarantine protocols will be followed as closely as possible without compromising the safety of the people and seals.

### ***Weather Conditions***

**Tsunami Warning or Other Evacuation:** In the event of a tsunami warning or other instance in which evacuation from KRF is necessary, the seals will be transported to an alternate site on Oahu where contamination from outside sources can be limited as much as possible. The seals will be loaded into cages and transported on a flatbed truck. Sprayers and shade will be provided to keep the seals cool. If deemed necessary by the veterinarian, the seals will be fed by throwing the fish in small basins of water on the floor their cages, or tube fed if not free feeding.

**Unusually hot weather:** Shade will be provided in each seal enclosure at all times. A rigid shade structure will be used rather than a tarp or shade cloth (noise created by wind on these soft coverings may frighten the seals). In unusually hot weather, running water will be provided to further cool the seals, either by turning up the pool inflow slightly to cause 1-2 inches of water to pool on the deck or by installing a sprinkler system that mists the seals' haul-out area with fresh water. Handling the seals in the middle of the day during periods of extremely hot conditions will not occur unless deemed necessary by the veterinary staff.

**Extreme winds:** All potential falling object hazards will be removed from around the seal enclosure. The enclosures will be monitored closely for hazardous debris that may have blown into the enclosure. Netting or equivalent structure will be placed over the pen and pool to prevent foreign objects falling into the seals' enclosure.

**Cold weather:** Waterproof-heating pads can be placed on the deck for the seals to lie on in instances of extreme cold or if a seal is otherwise not able to thermoregulate.

### ***Power Outages***

The pump that feeds water into the pools will not be operational during a power outage. For about 1 day the pool water can be maintained without an inflow source for 2-3 seals/pool (before bacterial levels reach unacceptable levels). To keep the pool water level with the deck so the seals can still haul out, both the outflow and overflow drains must be closed immediately. If the water level drops more than 8-12" below the deck, the seals will not be able to haul out. The drains can be closed after this point only if the fresh water source (hose water) is deemed clean enough to be used to fill the pool. If the pool becomes too dirty or the water level cannot be kept within 8-12" of the deck, the pool must be drained so the seals have access to a dry resting area. The seals will then be kept cool with shade and if appropriate, by a source of running freshwater.

A back-up generator supplies power to the main building, including the Vet Lab, and to 1 electrical outlet in the Fish House. Opening the freezers and refrigerators should be kept to a minimum to help these units maintain temperature during power outages.

***Potential Infectious Disease Outbreaks***

If one of the seals held in captive care exhibit signs of ill health and an infectious agent is the possible cause, the seal will be immediately moved to the designated isolation area. If it is unknown whether an infectious agent is the cause, the seal will be isolated and assumed infectious as a precautionary measure. Immediate sampling will be conducted following the Sampling Protocol (the “Re-check exam sampling protocol”) and every effort should be made to identify the causative agent. Treatment will be started before a diagnosis is made based on likely etiology as determined by the veterinary team.

- The NMFS Quarantine Policy will be followed.
- All equipment (feeding and cleaning supplies, handling gear, etc.) used to care for potentially infectious seals will be marked with a solid black triangle and will not be used to care for healthy seals. Isolation equipment will be stored separately and will not re-enter the clean animal area unless properly sanitized, if at all.
- Other seals exposed to a potentially infectious seal will remain separate from all unexposed seals (i.e. seals held in the same enclosure as the seal that became ill will not be mixed with those in a separate enclosure).
- There will be 1 animal care crew to care for the ill seals and another to care for healthy seals. Personnel caring for ill seals should not enter the clean seal area until showering and changing clothes.
- In each of the following locations, it is unlikely that a separate food preparation area will be available for the isolation area. If this is the case, a “one-way” fish kitchen will be set up in which contaminated items never enter the fish kitchen or healthy seal area. (For example, the amount of fish will be weighed out, thawed, and placed in a bag that will be handed to the sick animal caretaker at the base of the stairs to tanks 1 and 2.)
- The isolated seal will not be returned to its original holding tank until lab results verify the seal is healthy and the disease non-contagious.

Northwestern Hawaiian Islands: In the NWHI, the isolation area will be a separate pen erected off of the beach to prevent transmission of disease to other captive and wild seals. For example, on French Frigate Shoals, a quarantine pen will be erected at the southwest corner of the barracks on Tern Island if necessary.

Aboard the transport vessel: During vessel transport between the NWHI and Oahu, a separate pen will be erected on a lower deck of the ship if available or a separate area where cross-contamination is least likely (such as the bow versus the stern or the port and starboard sides). There will be sufficient space, more than 20 feet, and a solid barrier between the healthy animal and isolation areas to prevent direct contact, including splashed pool water and airborne disease transmission. If possible, there should be a separate footpath to the isolation area.

Kewalo Research Facility: See the Hawaiian Monk Seal Captive Care Protocol for Kewalo Research Facility, 2007-2008.



## **Papahānaumokuākea Marine National Monument Compliance Information Sheet**

**1. Updated list of personnel to be covered by permit. List all personnel names and their roles here (e.g. John Doe, Diver; Jane Doe, Field Technician, Jerry Doe, Medical Assistant):**

Rob Marshall, Field Biologist, Camp Leader  
Erin Pickett, Field Biologist  
Adam Fox, Field Biologist  
Artie Wong, Field Biologist  
Angie Kaufman, Field Biologist  
Chad Yoshinaga, Chief Scientist  
Charles Littman, Research Program Leader  
Colleen Siudsinski, Field Biologist  
Sarah Chinn, Field Biologist  
Julia Back, Field Biologist  
John Brack, Field Biologist  
TBN  
TBN  
TBN

**2. Specific Site Location(s): (Attach copies of specific collection locations):**

Deworming: will occur only on Laysan Island only and work will only be conducted on beach.

Collection for captive care could occur at any of the six main NWHI subpopulations (French Frigate Shoals, Laysan I., Lisianski I., Pearl and Hermes Reef, Midway Atoll and Kure Atoll), including any of the islets within the atolls.

**3. Other permits (list and attach documentation of all other related Federal or State permits):**

NMFS Office of Protected Species to take Hawaiian monk seals for Scientific Research and Enhancement Activities 10137-03 has been provided to PMNM already can send additional copy if needed.

**3a. For each of the permits listed, identify any permit violations or any permit that was suspended, amended, modified or revoked for cause. Explain the circumstances surrounding the violation or permit suspension, amendment, modification or revocation.**

None

**4. Funding sources (Attach copies of your budget, specific to proposed activities under this permit and include funding sources. See instructions for more information):**

Federal Agency Funded

**5. Time frame:**

Activity start: August 1, 2010

Activity completion: July 31, 2011

Dates actively inside the Monument:

From: August 1, 2010

To: July 31, 2011

Worming occurs on Laysan Island during our year round camp. Collecting and returning animals for captive care activities will be done intermittently on a TBD schedule based on the availability of charters, NOAA vessels and other modes of transporting animals.

Describe any limiting factors in declaring specific dates of the proposed activity at the time of application:

The deworming project is set and should occur on schedule upon receiving the permit.

Movement of seals for captive care activities will be done on an *as needed* basis when animals are identified as needing supplemental feeding or medical care and the necessary logistics will be put in place. The PMNM will be notified when such an activity is going to take place.

Personnel schedule in the Monument: August 1 2010 – July 31, 2010

**6. Indicate (with attached documentation) what insurance policies, bonding coverage, and/or financial resources are in place to pay for or reimburse the Monument trustees for the necessary search and rescue, evacuation, and/or removal of any or all persons covered by the permit from the Monument:**

As a U.S. Government agency, the project is self-insured. All personnel (contractors, state employees, and federal employees) are covered under workers' compensation.

**7. Check the appropriate box to indicate how personnel will enter the Monument:**

☒ Vessel

☒ Aircraft

Provide Vessel and Aircraft information:

Vessels:



Personnel and seals may be transported into/out of Monument on a variety of vessels: NOAA RV Oscar Elton Sette, M/V Searcher, M/V Kahana or others TBN (including potential Navy or Coast Guard support).

**Aircraft:**

Personnel and seals may be transported into/out of the monument via Midway on a number of aircrafts including the G4 currently operated by Chugach or using a Coast Guard C130.

**8. The certifications/inspections (below) must be completed prior to departure for vessels (and associated tenders) entering the Monument. Fill in scheduled date (attach documentation):**

- ☐ Rodent free, Date:
- ☐ Tender vessel, Date:
- ☐ Ballast water, Date:
- ☐ Gear/equipment, Date:
- ☐ Hull inspection, Date:

**9. Vessel information (NOTE: if you are traveling aboard a National Oceanic and Atmospheric Administration vessel, skip this question):**

**Vessel name:** TBN

**Vessel name:** The Searcher

Vessel owner: The Medical Foundation for the Study of the Environment

Captain's name: Captain Jonathan Littenberg

IMO#:

Vessel ID#:

Flag: USA

Vessel type:

Call sign:

Embarkation port: Kewalo Basin, Honolulu HI

Last port vessel will have been at prior to this embarkation: Honolulu HI

Length: 96 ft.

Gross tonnage: 230

Total ballast water capacity volume (m3):

Total number of ballast water tanks on ship:

Total fuel capacity: 9600 gal

Total number of fuel tanks on ship:

Marine Sanitation Device: Yes

Type: Type II USCG Approved

**Vessel name: MV KAHANA**

Vessel owner: KAHANA TUG LLC

Captain's name: MARIO LUIS

IMO#: 8108042

Vessel ID#: 643471

Flag: USA

Vessel type: SINGLE HULL OFF SHORE SUPPLY VESSEL

Call sign: WDC7062

Embarkation port: HONOLULU

Last port vessel will have been at prior to this embarkation: PORT ALLEN KAUAI

Length: 185'

Gross tonnage: 260 GRT

Total ballast water capacity volume (m3): 520 m3

Total number of ballast water tanks on ship: 7

Total fuel capacity: 62008 GAL

Total number of fuel tanks on ship: 6

Marine Sanitation Device: YES

Type: TYPE II USCG APPROVED

**Explain in detail how you will comply with the regulations regarding discharge in the Monument. Describe in detail. If applicable, attach schematics of the vessel's discharge and treatment systems:**

All overboard discharges are capable of being closed and the drains diverted to a dedicated holding tank that contains capacity to hold black and gray water for the duration of the cruise. The vessel can divert to outside the monument boundaries to empty the tank if necessary.

**Other fuel/hazardous materials to be carried on board and amounts:** No hazmat or fuel will be brought into the field specific for the deworming efforts. All hazmat and fuel is covered under the managers permit for the monk seal field camps.

Fuel will be brought in, stored on vessel for small boat operations to capture and transport seals.

**Provide proof of a National Oceanic and Atmospheric Administration (NOAA) Office of Law Enforcement-approved Vessel Monitoring System (VMS). Provide the name and contact information of the contractor responsible for installing the VMS system. Also describe VMS unit name and type:**

Kahana:

Thrane Sailor mini-C installed by Oceantronics

VMS Email: [436900529@c.xantic.net](mailto:436900529@c.xantic.net)

Inmarsat ID#: 436900529

Searcher: TBD

\* Individuals MUST ENSURE that a type-approved VMS unit is installed and that its automatic position reports are being properly received by the NOAA OLE system prior to the issuance of a permit. To make sure your VMS is properly configured for the NOAA OLE system, please contact NOAA OLE at (808) 203-2503 or (808) 203-2500.

\* PERMITS WILL NOT BE ISSUED TO INDIVIDUALS ENTERING THE MONUMENT VIA VESSEL UNTIL NOAA OLE HAS CONTACTED THE MONUMENT PERMIT COORDINATOR WITH A 'POSITIVE CHECK' READING.

#### **10. Tender information:**

On what workboats (tenders) will personnel, gear and materials be transported within the Monument? List the number of tenders/skiffs aboard and specific types of motors:

One 35 ft aluminum landing craft with inboard twin diesel engines; One 7m RHIB with inboard diesel engine.

One (1) Zodiac MK VI HD w/Yamaha 90 HP 4-stroke Outboard engine.

One (1) Zodiac MK IV HD w/Yamaha 50 HP 4-stroke Outboard engine

#### **Additional Information for Land Based Operations**

##### **11. Proposed movement of personnel, gear, materials, and, if applicable, samples:**

Movement within the monument will be accomplished using tenders of the Hawaiian Monk Seal Research Program. Samples/Seals will be moved utilizing the vessels/aircraft listed above.

**12. Room and board requirements on island:** NMFS staff will stay in program supplied tents. If a situation arises on French Frigate Shoals or Midway, NMFS will coordinate with USFWS for rooms to hold the minimal number of people to maintain seals prior to or after transport. This will be with as much advance notice as possible.

**13. Work space needs:** none

**DID YOU INCLUDE THESE?**

- ☐ Map(s) or GPS point(s) of Project Location(s), if applicable
- ☐ Funding Proposal(s)
- ☐ Funding and Award Documentation, if already received
- ☐ Documentation of Insurance, if already received
- ☐ Documentation of Inspections
- ☐ Documentation of all required Federal and State Permits or applications for permits

## **Appendix 1: Hawaiian Monk Seal Captive Care Protocol for Kewalo Research Facility, 2007-2008**

### **I. QUARANTINE**

#### **A. QUARANTINE DEFINITION AND OBJECTIVES**

1. Quarantine refers to “any isolation or restriction on travel or passage imposed to keep contagious diseases, insect pests, etc. from spreading (ref).”
2. Hawaiian monk seals currently held in captive care must be maintained under strict quarantine at all times to:
  - a. Minimize transmission of disease from outside sources- i.e. human contact
  - b. Minimize transmission of disease from captive care seals to susceptible animals, including wild seals, humans, etc.
  - c. Minimize transmission of disease among the three holding tanks at Kewalo Research Facility (KRF)
3. All personnel involved in the feeding, handling, and care of these seals must be properly trained in quarantine procedures by an experienced staff. (Quarantine procedures should always be posted in the food preparation area.)

#### **B. NMFS QUARANTINE POLICY**

##### **Quarantine from Outside Sources**

1. All equipment used in the quarantine facility, including feeding, handling, and medical supplies **MUST** be:
  - a. Labeled “MONK SEAL QUARANTINE”
  - b. Used exclusively for quarantined seals
  - c. Kept monk seal equipment separate from that used for other animals
  - d. Properly sanitized before and after entering the quarantine enclosures
2. **NO VISITORS** are allowed in monk seal quarantine area unless previous approval is granted by the on-site supervisor. Non-authorized personnel may be able to view the seals from an approved viewing platform outside the quarantine area.
3. Avoid direct contact with domestic or other captive or wild animals before and after entering Hawaiian monk seal quarantine enclosure. Shower and change clothes before and/ or after going to another animal care facility if entering the seal enclosures on the same day.
4. No street shoes are to be worn into the enclosures, including the walkway leading to tanks 1 and 2. Wear rubber boots/shoes designated for “monk seal quarantine” use in the enclosures at all times. Minimize wearing quarantined boots/shoes around KRF premises.
5. Dip soles of boots/shoes in dilute Nolvasan footbath upon entering **AND** leaving all the enclosures.
6. Immediately upon entering the enclosure to tanks 1 and 2, wash hands with antibacterial soap. Also wash hands before and after fish preparation, feeding, or handling seals. Always wash hands immediately after leaving a separate tank enclosure. Before and after entering tank 3, wash hands at sink next to the enclosure.
7. Any person that will potentially come in direct contact with quarantined seals for any procedure must wear sanitary protective clothing (i.e. coveralls, handling gloves, shoes) designated for quarantine monk seal use only. This clothing should be kept clean and in a designated area away from potential sources of contamination.
8. Protective clothing worn during procedures should be immediately washed in the washer with soap and dilute bleach solution following handling events.
9. Any new equipment or tools brought into the quarantine area must first be sanitized with a dilute bleach or Nolvasan solution.

### **Quarantine Between the Three Holding Tanks at KRF**

1. Separate equipment will be used to care for seals in each of the three tanks. This includes cleaning and feeding supplies (brooms, hoses, buckets, etc.) and handling gear (coveralls, booties, gloves). Keep this equipment separate.
2. A dilute Nolvasan footbath will be placed at the door of each tank to be stepped before and after leaving the enclosure. A freshwater bath will be placed as a final rinse before entering the pen.
3. Personnel must change protective clothing when caring for seals housed in tanks 1 and 2. However, personnel caring for isolated seals in tank 3 are prohibited from entering the "healthy" seal area (the stairs, walkway, fish house, and tanks 1 and 2).
4. Seals housed in separate enclosures will not be mixed unless deemed necessary by the veterinary staff.
5. A seal require isolation, follow the Potential Disease Outbreak Protocol.

## **II. OBSERVATIONS AND CONDUCT AROUND SEALS**

### **A. OBSERVATIONS OF THE SEALS**

1. In the morning and prior to each feed, conduct a thorough inspection of the seals and pens before proceeding with further activity. Following each feed or handling event, monitor the seals' behavior closely. Perform a final inspection before leaving for the day.
2. Throughout the day monitor and record the behavior of each seal. Observe the condition and activity level of the seals and presence of feces, urine, spew, and harmful debris in or around pens. When possible, note ID of seal that produced scat, spew, etc. Note the color, consistency, and amount of scat, urine, and spew.
3. Note anything unusual in a seal's normal appearance (eyes, nasal discharge, bite wounds, etc.) and behavior (lethargic, unresponsive, etc.). Notify attending veterinarian and animal care manager immediately of any abnormal changes in a seal's health.
4. Succinctly record any observations on the "Observation" form in each seal's chart, including the time and observer's initials. Frequently used acronyms: BAR = bright, alert, and responsive; QAR = quiet, alert, and responsive.

### **B. CONDUCT AROUND THE SEALS AT ALL TIMES**

Every possible effort should be made to minimize the habituation of the seals by reducing human-seal interactions.

1. When in enclosures, **DO NOT MAKE PHYSICAL CONTACT WITH SEALS** unless necessary for procedures requiring handling. Minimize going into the enclosure and the amount of time you spend in the enclosure as much as possible.
2. If seals are resting or sleeping, do not make loud noises or startling gestures, and move slowly when in close proximity to them to minimize stress.
3. Minimize talking when working with or near the seals and the enclosure.
4. Whenever possible, observers should remain as inconspicuous and unobtrusive as possible to observe seals' normal behaviors in captivity and minimize their stress in captivity.
5. Each person entering an enclosure with the seal should be carrying a herding board, which should be within arms-reach at all times.
6. Outside of feeding sessions seals may display undesirable behaviors which include: a) approaching too closely or too rapidly; b) mouthing hoses, brooms, or boots; and c) stereotypic behaviors which include repetitive splashing or slapping at the walls of the enclosure. If seals approach too closely or too rapidly use a herding board to keep the seal away. The mouthing of brooms, hoses, and boots should be discouraged by preventing opportunities for seals to bite at these objects in the first place. Stereotypic behaviors are a sign of boredom and may be reduced by providing seals with their approved environmental enrichment devices (EEDs).

### **III. CLEANING THE QUARANTINE AREA**

#### **A. DISHES**

1. Wash all dishes used for feeding and handling with dish soap and water. Rinse thoroughly.
2. Soak all metal and rubber equipment (bolus syringes, knives, tongs, etc.) in dilute Nolvasan for at least 10 minutes.
3. Soak all plastic equipment (cutting boards, buckets, cooler, etc.) in dilute bleach for at least 10 minutes.
4. Rinse all dishes thoroughly to remove the dilute bleach or Nolvasan.
5. Allow all dishes to air-dry.
6. Stomach tubes should be washed with soap and water, rinsed thoroughly, and then boiled for 10 minutes. Be sure to scrub the inside of the feeding tubes. Keep sanitized stomach tubes wrapped in a clean towel.
7. Bolus Syringe Care: after the syringes have been washed and dried as described above, lubricate the O-ring with mineral oil and put the syringes back together for safe storage. Be careful when handling the syringes as they are fragile and can crack easily.

#### **B. DAILY CLEANING AND MAINTENANCE**

##### **Seal Enclosure Cleaning**

Do not allow seals to mouth or bite brooms or fresh water hoses. If the hose enters the pool remove it immediately. Never allow the broom, hose, or any equipment to remain unattended in a seal enclosure. Return all equipment to its storage area after use (i.e. coil and hang hose). Always keep the enclosure doors securely bolted because the seals are very adept at exiting the enclosures through a door left ajar. When cleaning, take the opportunity to inspect urine for color and feces for consistency and parasites. Always record feces and urine in the observations form in the seal's chart and make special note of any unusual findings.

1. After the morning feed, the entire pen enclosure should be checked for any scat, urine, fish parts, and wind-blown debris. If necessary, use a broom and fresh water hose to clean the seal enclosure. Thoroughly rinse all fish scales, blood, and debris from the decks, walls, and ledge of the enclosure and walkway with the fresh water hose after each feed. Special care should be taken to clean scales from doors, door handles, and bolts.
2. Before leaving in the evening, the deck and pool walls and floor should be hosed down and any spattered blood, scales, scat, or other debris should be scrubbed away.

##### **Miscellaneous Cleaning**

1. Rinse off the walkway and stairs leading to the seal enclosure at least once a day. Scrub the walkway with broom and water as needed.
2. Refill footbaths 1-2 times a day or as needed with dilute Nolvasan (usually once first thing in the morning is fine). Add 3 oz Nolvasan to 1 gallon water. Be sure to have a final water rinse before the pen entrance. Old dilute Nolvasan should be poured onto the pavement next to and at the base of the stairs (don't dump it down the stairs).

##### **Fish house Cleaning**

1. Freezers and refrigerators must remain clean and neat at all times. All feeders are responsible for maintaining freezer cleanliness on a daily basis. Keep freezers free of ice buildup as much as possible.
2. Wipe down all counter and table surfaces after each feeding. Be especially mindfully of cleaning any fish scales and spattered blood from the all surfaces after each feeding.

3. Mop the fish house floor with a dilute bleach solution (1 part bleach to 30 parts water) after the morning feeding.
4. Empty the garbage and take it to the outside dumpster at the end of each day (or every other day).
5. All damaged or unused fish and fish parts including the scales should be put in the "Fish Waste" bag in the chest freezer and should NOT be thrown away in the regular trash. The "Fish Waste" bag should be taken to the facility dumpster every Wednesday before 0800 for immediate pick-up.

#### **Coverall Cleaning**

1. Wash all coveralls, kneepads, gloves, and booties following each use with dilute bleach and laundry detergent in the washing machine at the end of each day. Do not set quarantine items down outside the quarantine area.
2. Dry all items in the dryer except the booties with rubber soles. The booties should be air-dried on the floor in the fish house.
3. In between handling events on the same day, hang the coveralls in the sun to dry.
4. Store clean, dry coveralls, etc. in the box in the fish house labeled "Clean Handling Equipment" (in the cabinet).

### **C. WEEKLY CLEANING**

#### **Cleaning Seal Enclosure**

The monk seal tanks and enclosures are drained and cleaned once a week. A minimum of 3 people should conduct the weekly cleaning. Pool cleaning occurs every Friday following the 0800 feed. A dilute bleach solution should be used (the black sprayers holds 2 gallons so add about 1 cup of concentrated bleach). When using bleach solutions always direct the rinse water away from seals because the bleach solution is a skin and eye irritant. Use the large, soft-bristled brushes for all cleaning.

1. Empty all Nolvasan footbaths except for one at the base of the stairs. Once the footbaths are emptied, minimize leaving and reentering the quarantine area as much as possible.
2. Spray the walkway with the dilute bleach mixture. Be sure to spray the walls, ledge, and doors. Direct the bleach spray away from the seal enclosure. Scrub the walkway deck and ledge. Let the bleach stand for 10 minutes.
3. Hose off the dilute bleach thoroughly – perform at least 2 washes of all surfaces.
4. When you are convinced that all the bleach has been rinsed away, move the seals into the holding area. Use a net to scoop the seals out of the water and herding boards to direct them into the holding area. Be sure to keep the seals cool with running water while cleaning the enclosure and monitor the seal's affect and behavior regularly.
5. Once the seals are secure in the holding area, begin draining the pool and start bleaching the deck. Spray the entire deck and up to the fiberglass line on the walls. Be careful to avoid sending bleach (from the sprayer or wind) into the seals' holding area. Spray the cages, any enrichment tools, and drain covers. Scrub the deck floor and allow the bleach to stand for 10 minutes.
6. As the pool continues to drain, thoroughly rinse the bleach from the deck area. Perform at least 2 rinses of all surfaces.
7. When the pool is drained, scrub the walls and floor of the pool to remove all scales and spattered blood. Using the hand-held brushes works well for scrubbing the walls.
8. Spray the pool walls and floor with dilute bleach and scrub all surfaces again. Let the bleach stand for 10 minutes.
9. Rinse the bleach from the pool walls (at least twice) while simultaneously turning on the water inflow. Leave the bottom drain open, with the drain cover in place, for several minutes to thoroughly rinse all the bleach down the drain. When you feel confident that all the bleach has been washed away, close the bottom drain, and begin filling the pool.



10. Flush the deck for several minutes to remove any bleach remnants. Remove all cleaning equipment then bring the water level with the deck and re-introduce the seals to the enclosure.
11. After all the tanks and walkways have been cleaned, thoroughly rinse the bleach solution from the brooms and all cleaning equipment. Separate cleaning supplies are used in each of the tanks. Replace the Nolvasan solution (3 oz/1 gal) in all footbaths.
12. Record the seals' behavior, the duration spent in the holding area, and any other relevant information from the cleaning event (scat, spew, urine, etc.) on the observations form in each seal's chart.

#### **IV. WATER SAMPLING SEAL TANK**

Sampling should occur on the same day (Tuesday) and time (1000-1100) each week at least a couple of days after the weekly enclosure cleaning. We collect one sample from the pool and one from the inflow in addition to a temperature control sample collected from the pool. These samples are submitted to Hawaii Food & Water Testing Lab (HF&WTL) for fecal coliform testing.

1. Try to be as sterile as possible: wear gloves, do not open lid to bottle until immediately before collection, do not contaminate inside of lid or bottle, don't set the lid down, etc.
2. Collect the inflow sample by removing the lid and holding the bottle under the water inflow to fill it. Decant any excess water being careful not to touch the lip of the bottle or the lid.
3. Sample the pool (pool and temp control sample) 180° from the water inlet. With the lid still in place, submerge the bottle about 1 foot deep. Unscrew the lid underwater with the bottle positioned counter-current to fill the bottle. Replace the lid underwater. Remove the bottle from the water and decant the excess water being careful not to contaminate the bottle or lid.
4. Immediately place the samples in the small red cooler with blue ice (provided by HF&WTL) for transport to the lab. If transport is not immediate, place the samples in the refrigerator (sampling fridge, not fish storage fridge). Store sample bottles in the cooler and ice pack in freezer until next sampling.
5. Complete all the necessary paperwork and be sure to label each bottle (pool, inflow, temp control).
6. Results submitted on Tuesday are usually faxed to us, c/o Robert Dollar, on Thursday or Friday. These counts should not exceed 1000 MF/100ml. If fecal coliform counts exceed 1000 MF/100ml, results are reported to Robert Dollar by phone; sampling must be repeated within 24 hours. Promptly notify the veterinary staff if counts are above 1000 MF/100ml.
7. Enter the date, time, coliform count, and any pertinent comments in the HMS Water Testing spreadsheet.

#### **DIRECTIONS TO HF&WTL**

2688 B Kilihau St.  
Honolulu, HI 96819  
Ph: 836-5558  
Fax: 836-5509  
contact: Wendy

Open Mon.-Friday, 8am-5pm

Located in Mapunapuna near the airport. Go west (towards the airport) on Nimitz Hwy & turn right on Kakoi St then right again on Kilihau St (2688B Kilihau St.). It's the 3<sup>rd</sup> grey building on the left.

#### **V. SEAL ILLNESS/EMERGENCY CARE**

1. In case of an emergency or suspected illness, refer to the phone list and call the attending veterinarian or veterinary technician immediately to relate symptoms or circumstances of emergency or illness. Follow the emergency chain-of-command protocol.
2. A veterinarian or trained veterinary staff will perform any needed blood sampling.

A crash kit and emergency drugs are kept in the fish kitchen. All other medical supplies for blood sampling, fluid and antibiotic administration, monk seal medications, and additional medical supplies are kept at

## Appendix 2: Hawaiian Monk Seal Feeding Protocol

- Begin tube feeding the seal at the next scheduled feeding time after admit or as determined by the veterinary staff.
- Weaners: Feed the seals three times per day with 5-6 hours between each feed.
- Yearlings: Tubings may be reduced to twice a day to reduce stress, if determined by the veterinarian.
- The seals will be tube feed until they begin eating dead herring on their own.
- All seals will be re-hydrated with electrolyte fluids for the first 24 hours. After that time, a diluted fish mash slurry will be introduced followed by full-strength fish mash. The volume to be tubed is determined by admit weight as described below.
- Begin offering yearlings fish prior to tube feedings 36 hours after admit.
- Begin offering weaners fish prior to tube feedings 72 hours after admit.
- Initially offer 0.25 kg of fish at each scheduled feed before attempting to tube feed. If the seal does not eat the entire amount of fish offered, then tube feed according to the weight specifications.
- Once the seal has consistently eaten 0.25 kg of fish at 3 feeds, the seal is now considered a free feeder. Begin increasing the feed amount by 0.25 kg every day at each feed until a daily intake of about 6-7% the seal's body weight is reached.
- After the first 72 hours, if a seal is not eating 2 days later, begin increasing the tubing volume by 50 mL every day at each feed. The maximum tubing volume will be determined by the seal's weight and response to the increased volume at each tubing.

### Feeding Schedule for First 72 Hours

Feed #	Admit Weight				Notes
	< 30 kg	30-39 kg	40-50 kg	> 50 kg	
1	300 mL EL	500 mL EL	750 mL EL	1000 mL EL	
2	300 mL EL	500 mL EL	750 mL EL	1000 mL EL	
3	300 mL EL	500 mL EL	750 mL EL	1000 mL EL	
4	150 mL EL 150 mL FM	250 mL EL 250 mL FM	375 mL EL 375 mL FM	500 mL EL 500 mL FM	
5	150 mL EL 150 mL FM	250 mL EL 250 mL FM	375 mL EL 375 mL FM	500 mL EL 500 mL FM	
6	150 mL EL 150 mL FM	250 mL EL 250 mL FM	375 mL EL 375 mL FM	500 mL EL 500 mL FM	Yearlings: begin offering fish, tube if not eating
7	300 mL FM	500 mL FM	750 mL FM	1000 mL FM	
8	300 mL FM	500 mL FM	750 mL FM	1000 mL FM	
9	300 mL FM	500 mL FM	750 mL FM	1000 mL FM	Weaners: begin offering fish, tube if not eating

EL = Electrolytes  
 FM = Fish Mash

### ELECTROLYTE (EL) SOLUTION

A soluble powder electrolyte supplement (BounceBack®, Manna Pro, St. Louis, Missouri) will be used to help the seals retain essential body fluids.

1. Mix 1 scoop of BounceBack with 2 quarts of filtered water.
2. Stir until the powder is completely dissolved.
3. When finished, pour the solution into a clean container and label it with “electrolytes” and the date, time and your initials.
4. Store in the refrigerator for up to 24 hours.

### **FISH MASH (FM) PREPARATION**

Fish mash is equal parts fish and filtered water blended together using a commercial-strength blender. For example, you would weigh out 1 kg of chopped fish and add 1000mL of water. About 2 batches of fish mash can fit in the blender at a time. More concentrated fish mash can be made or additional sources of fat (salmon oil) may be added if necessary as directed by veterinary staff.

1. Cut off the heads, fins, and tails from slightly defrosted fish. Chop the fish into small pieces.
2. Weigh out 1 kg of fish pieces.
3. To blend, add 1000mL of water and about half the amount of chopped fish pieces. Make sure the fish is not too frozen as this will wear out the motor on the blender. Do not run the blender or more than one minute at a time.
4. Blend on low for 10 seconds.
5. Add the rest of the fish pieces.
6. Blend on low for 10 seconds then on medium for 30-45 seconds.
7. Stir the fish mash with spatula to make sure there are no large chunks. If there are any chunks that might get stuck in the stomach tubes, blend again.
8. When finished, pour the fish mash into a clean container and label it with the type of formula, date, time and your initials.
9. Store in the refrigerator for up to 24 hours.

### **FISH PREPARATION**

IQF herring is stored in the chest freezer in the Fish House and thawed on an as-needed basis. All frozen fish must be well sealed to prevent freezer burn and drying. Always re-tie the plastic bags of herring after each use. Any fish that are broken or “damaged” should not be used.

1. Review feed amounts in each seal's chart before preparing fish. This information is also posted on the whiteboard in the fish house.  
Note: Check the orders form for instruction on fish prep for each seal. For example, you may need to cut off the head and pectoral fins (on a diagonal), the anal and dorsal fins, and tail.
2. Wash hands thoroughly with antibacterial disinfectant soap before handling buckets and fish. Always wear gloves when preparing feeds.
3. Thaw the fish in running water. Make sure the drain covers are in place so scales don't clog up the pipes. While thawing, inspect the fish again for bruises, cuts, or other abnormalities not detectable when frozen. Discard any damaged or broken fish.
4. Throw accumulating scales from the drain covers and all unusable fish parts in the plastic bag in the chest freezer labeled “Fish Waste.” These will be put in the facility dumpster just prior to trash pick-up early Wednesday morning.
5. Weigh the fish using the digital scale and each seal's feeding pail. Turn the scale “ON” and allow it to zero. Put the pail on the scale and let it equilibrate, then push the “TARE” button. Tare the scale before each seal's feed. Record the feed weights in the feeding log in each seal's chart.
6. If supplementing the herring feed with a multivitamin (Pinnivite = PV) insert the vitamin tablet into the gill slit or body cavity of one of the fish.

7. If not feeding immediately, keep the fish cool in the refrigerator or on blue ice in a cooler until ready to feed. Keep feed amounts for each seal separate.
8. Fish can be defrosted in the refrigerator prior to use but once thawed, the fish are only good for a maximum of 24 hours in the refrigerator. Thawed fish should be labeled with the date and time that the fish was defrosted.

### **TUBE FEEDING**

Various methods can be used to tube feed a seal. Use the method that the feeder and restrainers are most comfortable with, decided on before entering the seal enclosure. The minimal number of people should be used to restrain the seal.

1. Wash hands with disinfectant soap immediately prior to and after feeding.
2. Prepare the electrolyte solution, fish mash, or other formula (check in the refrigerator as there may be some already made within the last 24 hours).
3. Measure out the specified amount according to the seal's chart (measure out a little extra to account for fish mash lost before the tubing). This information is also posted on the whiteboard in the fish house. Return any unused electrolytes/fish mash to the refrigerator.
4. Warm up the electrolytes/fish mash for 30 seconds in the microwave. Thoroughly stir the fish mash to distribute the heat. A second round of warming may be required to remove the chill (be careful not overheat).
5. Ground up the vitamin and any other supplements or medications with a mortar and pestle. Pour this into the electrolytes/fish mash and stir thoroughly.
6. Draw up the electrolytes/fish mash into 1-2 500 mL bolus syringes.
7. Put a feeding tube, the 1-2 bolus syringes (keep upright), and any remaining electrolytes/ fish mash (in a beaker) for the feed into a bucket to be taken into the seal enclosure.
8. Anyone entering the enclosure to handle the seals should be wearing gloves, coveralls, and booties and step in Nolvasan footbath and freshwater rinse before entering pen. Use a separate set of coveralls when working in each seal enclosure. Have a clear plan of action before entering the pen.
9. Keep the seal cool during and post-handling. Hose off post-handling if necessary. Restrain and feed in the shade if possible.
10. Enter the enclosure slowly and minimize startling the seals.
11. Once the restrainer(s) have the seal under control (use a towel and herding boards), the feeder inserts the tube into the mouth of the seal. The head restrainer may need to "pop the jaw" by inserting his/her thumbs or forefinger between the jaw behind the molars (outside the mouth).
12. Slowly insert the tube into the stomach, allowing the seal to breathe normally.
13. Ensure that the tube is in the stomach before administering the electrolytes/fish mash by listening for stomach gurgling. Blowing gently on the end of the tube may be required to hear these sounds.
14. Gently and steadily plunge the electrolytes/fish mash into the stomach. Crimp the tube when removing a bolus syringe from the stomach tube so as not to create a vacuum. The stomach tubes hold about 50 mL so after administering all the electrolytes/fish mash, plunge about 50 mL of air through the tube.
15. Slowly remove the stomach tube.
16. After the tube feeding, release the seal and exit the enclosure immediately. Remove all feeding equipment. From outside the pen, watch closely for regurgitation, scat, and post-handling behavior.
17. Record observations associated with the feeding and handling in each seal's chart (in the Observation form) and record the amount feed in the Dietary/Medical Record form.

18. Perform all post-feeding cleaning duties including: washes all dishes, cleaning all surfaces in the fish house, and mopping the fish house floor (morning feed only).

### **OFFERS AND FISH SCHOOL**

There are a variety of techniques that can be used to encourage the seals to begin free feeding as quickly as possible, which are outlined in Guidelines for Offers and Fish School.

### **FREE FEEDING**

1. Wash hands with disinfectant soap immediately prior to and after feeding.
2. From the walkway between tanks 1 and 2, throw fish to each seal at a rapid pace to minimize begging behavior. Throw a handful of fish at one time to always have at least a couple of fish waiting in the water for the seal to eat so that the seals are looking in the pool for fish. Don't throw a fish when the seals are looking or vocalizing at you. Try to keep the seals on opposite sides of the pool as much as possible. It is preferable that each seal eat only the fish from their bucket but if they steal each other's fish, swap fish of similar size from the other's bucket to make sure each eats their share.
3. Give the vitamin fish after observing that the fish are being eaten (second or third fish). Closely observe if vitamins are ingested or sink to bottom of the pool. If a seal does not receive vitamins, record this in the seal's chart (on the dietary/medical record and observations forms) and leave a note that they should be given at the following feed. Notify the veterinary staff ASAP if medications are not received.
4. Any uneaten fish should be disposed of in the "Fish Waste" bucket in the chest freezer. Scooping the pool for uneaten fish pieces is often unnecessary but if large chunks of fish are not consumed, scoop these from the pool and weigh this amount (subtract from the amount fed when recording in dietary record form). Be sure to clean, bleach, and thoroughly rinse the net after each use.
5. Record observations associated with the feeding in each seal's chart, including any adverse behavior (on the Observations form). Record how much each seal actually ate, estimating weight of additional fish ingested or fish lost when necessary, in the Dietary/Medical Record form. Frequently used acronyms: AWIP = ate well in pool.
6. Perform all post-feeding cleaning duties including: hosing off splattered blood and scales, washing all dishes, cleaning all surfaces in the fish house, and mopping the fish house floor (morning feed only).

### **Appendix 3: Hawaiian Monk Seal Medical Care of the Seals**

The medical treatment regime for a sick or injured seal will be determined on a case-by-case basis by the veterinary staff. Detailed observations by caregivers will greatly facilitate medical decision-making. The following factors will be considered to help determine the appropriate treatment regime for a seal:

- Overall attitude – activity level and behavior (lethargic, depressed, quiet, lying curled up, hunched posture, logging in the water, active, alert, interacting with other seals, swimming, hauled out, etc.)
- Presence of vomit, urine, or feces (amount, color, and consistency)
- Appetite and feeding behavior (loss of appetite, no interest in fish, loss of interest abrupt or gradual)
- Experience with seals with similar clinical signs (with Hawaiian monk seals and other pinnipeds handled at TMMC)

#### **Daily Medical/Physical Examination**

A complete medical/physical examination will be performed daily using the Physical/Medical Exam Form to record data. Seals WILL NOT be restrained for this physical exam; it will be strictly a visual exam. If any abnormalities are found, notify the veterinary staff immediately. A more thorough description of any abnormalities should be documented in the Medical Progress Report by the veterinary staff. Also complete the Wound Sheet or Eye Exam Form if applicable. Photographs of any physical abnormalities will also be taken. The course of treatment and sample collection of all abnormalities will be determined by a committee of veterinarians and experienced monk seal caretakers.

#### **Frequently Encountered Medical Problems and Treatments**

##### **Malnutrition**

For all underweight seals, the objective is to maintain a positive energy balance and good hydration status. A sign of good hydration is tearing eyes (wet circles around the eyes). Weekly weights will be used to assess if the caloric needs of each seal are being met. Severely underweight seals may be too weak to fight infections or other illness so all efforts will be made to eliminate any factors that might compromise the health of the seal, most of which are standard operating procedures: limiting exposure to the elements (both heat and cold), reducing the stress on the seal, maintaining quarantine.

- Follow the appropriate feeding protocol.
- It may be necessary to use a formula with a higher caloric value (see Feeding Protocol) or increase the number of feedings per day.
- Subcutaneous fluids with dextrose may be administered to provide additional calories.
- If a seal is still not gaining weight although eating an adequate number of calories, check the blood work to investigate potential underlying causes.
- Anthelmintics, which may improve absorption, will be administered to reduce the load of gastrointestinal parasites when the seal is hydrated.

##### **Stress**

During past captive care efforts with Hawaiian monk seals, some seals, especially yearlings and other older seals, have exhibited signs of acute stress. Clinical signs of stress include stereotypic behavior, which is classified as any repetitive motor behavior that does not have an obvious purpose or function. Examples of stereotypic behaviors include pacing and swimming in circles. Stress may inhibit the seal's

ability to fight underlying infections or illnesses; every effort should be made to reduce the stress of the seal and support the seal nutritionally.

- Maintain a positive energy balance and good hydration status as outlined above.
- Maintain normal body temperature (protect from the elements)
- In extreme cases, the activity level of the seal may need to be limited by minimizing or restricting access to the swim area. This approach will be used very cautiously so that it does not further stress the seal.
- A change in the group size or structure may be tested for its effects on seal stress behavior.
- Medication to reduce the stress level of the seal may be administered as prescribed by the attending veterinarian. Such medications include diazepam and haloperidol.

### **Periods of Inappetence**

Once free feeding, a seal may stop eating at any time. Satiation, warm weather, stress, or ill health may cause inappetence. If satiation is the cause, the seal should quickly resume free feeding after cutting its dietary intake for a brief period (each seal has a different satiation point; the feed orders will be adjusted for each seal so as not to exceed this limit).

- If a seal does not eat at 1 feeding session, monitor the behavior and overall affect of the seal. At the next feed, offer the normal feed amount. Monitor the feeding behavior closely during this feeding session.
- If the seal is still not eating after 2-3 feeds, complete a medical examination, draw blood and tube electrolytes at the same time (tube volume according to weight specifications). It is important to re-hydrate the seal with several electrolyte tubings before returning to offering fish.
- Subcutaneous fluids may also be administered as determined by the veterinary staff to help the seal re-hydrate more quickly.
- A rectal swab (Cary Blair media) or fecal sample may be collected if a gastrointestinal bacterial infection is suspected as the cause of the inappetence.
- Proceed with offers before tube feedings using an abbreviated 72-hour feeding schedule as determined by the veterinary staff (may increase the tubing/offer amount more rapidly).

### **Multiple vomiting episodes**

Vomiting episodes may accompany periods of inappetence. If spew is discovered in the pen, make every effort to identify the seal that produced the spew. Monitor all the seals in the pen at subsequent feedings for any signs of inappetence to help identify this seal.

- For an isolated instance of vomiting, monitor the appetite and behavior of the seal. Carefully check for additional spew.
- If multiple vomiting episodes occur, stop feeding the seal for 12 hours. Administer 1 shot of DualPen (1 ml/10kg IM) during the first 12 hours of not eating to treat for possible Clostridium infection. If signs do not resolve, draw blood and begin a 7-day treatment of DualPen (1 ml/10kg IM every other day).
- Support the seal nutritionally with tube feedings until it is eating again.
- A rectal swab (in Cary Blair media) or fecal sample may be collected if a gastrointestinal bacterial infection is suspected as the cause of the vomiting.
- Other medications and subcutaneous fluids may be administered as determined by the veterinary staff (such as Pepto-Bismol, cimetidine, metaclopramide).

### **Abscesses**

Seals may be admitted with abscesses or may develop them while in our care, usually from bites by other seals. Treatment depends on the site and severity of the abscess.



- Lance the abscess with a sterile scalpel blade, swab the inside of the abscess with a culturette and place in Amies transport media, and flush with dilute chlorohexadine or hydrogen peroxide every other day until no more pus is visible at the drainage site.
- For small abscesses (quarter-sized or smaller), antibiotics or other medications will not be administered if the seal is acting healthy.
- For large abscesses, treat with antibiotics, typically amoxicillin, according to the dosages on the Drug Formulary.
- If the abscess recurs, check the culture and sensitivity results of the initial swab for choice of antibiotic treatment.

### **Eye lesions**

The eyes will be closely monitored during the daily medical/physical examination.

- If any signs of ocular disease develop, start daily ocular exams using the Eye Exam Form to record data.
- Four swabs of the cornea and conjunctiva will be collected. One swab will be used for bacterial culture, one for herpes PCR, the others banked at -70°C.
- Topical antiviral and antibiotic treatment will be started immediately according to the Eye Disease Protocol
- Isolate the seal(s) with ocular diseases and maintain strict quarantine.
- Commence rounds with a veterinary team to monitor treatment and progress. Frequency of rounds will vary, but will be at least once a week.

#### **Appendix 4: Hawaiian Monk Seal Sampling Protocol**

The Sampling and Morphometric Checklist Form must be completed for all admit, re-check, and release exams.

##### *Admit Exam*

All seals collected for inclusion in captive care will undergo an admit exam 1-3 days after being collected. During this exam, the seal will be physically restrained and a blood sample, fecal sample, 2 swabs of each orifice (eye, nasal, oral, rectal, and genital), standard length, axillary girth, and body weight will be collected.

##### *Re-check Exams*

- If a seal is clinically healthy and has normal blood work (values within normal ranges for Hawaiian monk seals), blood will be drawn and re-checked every 4 weeks (to make sure blood work is still normal and to bank a series of control samples for each seal).
- If a seal has abnormal blood work, blood will be drawn and re-checked every 1-2 weeks until normal levels are attained.
- If a seal is showing signs of ill-health, obtain a blood sample as soon as possible and re-check every 1-2 weeks until normal levels are attained.
- If any abnormalities are present, additional blood sampling may be requested by the veterinarians. Other samples (swabs, biopsies, etc.) may also be requested by the veterinarians.

##### *Release Exam*

All seals included in captive care will undergo a release exam 4-7 days prior to release. During this exam, the seal will be physically restrained and a blood sample, fecal sample, 2 swabs in each orifice (eye, nasal, oral, rectal, and genital), blubber biopsy, standard length, axillary girth, and body weight will be collected.

##### *Blood sampling*

- Collect up to 10 mL of whole blood from the extradural venous sinus during each exam. A larger volume of blood may be collected once the seal is hydrated, at the release exam, or as determined necessary by the veterinary staff for diagnostic purposes.
- Collect 1 EDTA lavender top tube (LTT) for CBC. Fill the LTT half way and immediately but gently invert 5-6 times to mix additive with blood (ensure the blood is not clotted). After CBC testing is complete, spin the tube in a centrifuge for 10 minutes and pipet off the plasma and bank (label PS LT). Also pipet off and bank the buffy coat (label BU LT).
- Collect at least 2 serum separator tubes (SST) for serum chemistry and serum banking. Do not invert SST's. Let SST tubes sit at room temperature for at least 15-30 minutes (but not more than 1 hour); then spin tubes in centrifuge for 10 minutes. Pour off the serum into 1 mL aliquots (label SE).
- Collect 1 heparin green top tube (GTT) for serum chemistry testing using an I-Stat (for immediate estimates of blood values). Fill the GTT half way and immediately but gently invert 5-6 times to mix additive with the blood (ensure the blood is not clotted). After I-Stat testing is complete, invert the GTT 5-6 times to re-mix and pipet off 0.1 mL blood into a cryovial to be banked for biotoxin testing. Also apply a drop to each of the eight circles on the biotoxin card (see NMFS Hawaiian Monk Seal Sampling Protocol). Spin the tube in a centrifuge for 10 minutes and pipet off the plasma and bank (label PS GT). Also pipet off and bank the buffy coat (label BU GT).

- All banked samples should also be labeled with the seal's permanent ID, date collected, and specimen #.
- Complete Blood Count (CBC) testing: white blood cell count, white blood cell differentials, red cell count, hemoglobin, hematocrit, mean corpuscular volume, mean corpuscular hemoglobin, mean corpuscular hemoglobin concentration, platelet count
- Serum chemistry testing: sodium, potassium, chloride, blood urea nitrogen, creatinine, glucose, calcium, phosphorus, uric acid, total protein, albumin, globulin, osmolarity, total/direct/indirect bilirubin, gamma glutamyl transpeptidase, lactate dehydrogenase, aspartate aminotransferase (serum glutamic-oxaloacetic transaminase), alanine aminotransferase (serum glutamic-pyruvic transaminase), alkaline phosphatase, iron, cholesterol, triglycerides, creatine phosphokinase, amylase, lipase, fibrinogen

#### *Fecal examination*

- One fecal sample will be examined for presence of parasite ova by fecal float. Samples can be held at 4°C if examined within one week of collection, otherwise they should be preserved by addition of equal volume of formalin to the fecal container.
- Aerobic culture of feces will be done on admit, release and any diarrhea samples as directed by the veterinarian. Swabs for fecal culture should be placed in Cary Blair transport media and stored at 4°C if cultured within one week of collection, otherwise frozen at -70°C until culture is possible.

#### *Swabs*

- Swabs of each orifice should be stored in dry cryovials at -70 C

## **Appendix 5: Contingency Plans for Hawaiian Monk Seals Held at Kewalo Research Facility**

As with an emergency situation, the safety of the personnel will be the first priority followed by the safety and health of the seals. Animal care and quarantine protocols will be followed as closely as possible without compromising the safety of the people and seals.

### ***Weather Conditions***

Tsunami Warning or Other Evacuation: In the event of a tsunami warning or other instance in which evacuation from KRF is necessary, the seals will be transported to an alternate site on Oahu where contamination from outside sources can be limited as much as possible. The seals will be loaded into cages and transported on a flatbed truck. Sprayers and shade will be provided to keep the seals cool. If deemed necessary by the veterinarian, the seals will be fed by throwing the fish in small basins of water on the floor their cages, or tube fed if not free feeding.

Unusually hot weather: Shade will be provided in each seal enclosure at all times. A rigid shade structure will be used rather than a tarp or shade cloth (noise created by wind on these soft coverings may frighten the seals). In unusually hot weather, running water will be provided to further cool the seals, either by turning up the pool inflow slightly to cause 1-2 inches of water to pool on the deck or by installing a sprinkler system that mists the seals' haul-out area with fresh water. Handling the seals in the middle of the day during periods of extremely hot conditions will not occur unless deemed necessary by the veterinary staff.

Extreme winds: All potential falling object hazards will be removed from around the seal enclosure. The enclosures will be monitored closely for hazardous debris that may have blown into the enclosure. Netting or equivalent structure will be placed over the pen and pool to prevent foreign objects falling into the seals' enclosure.

Cold weather: Waterproof-heating pads can be placed on the deck for the seals to lie on in instances of extreme cold or if a seal is otherwise not able to thermoregulate.

### ***Power Outages***

The pump that feeds water into the pools will not be operational during a power outage. For about 1 day the pool water can be maintained without an inflow source for 2-3 seals/pool (before bacterial levels reach unacceptable levels). To keep the pool water level with the deck so the seals can still haul out, both the outflow and overflow drains must be closed immediately. If the water level drops more than 8-12" below the deck, the seals will not be able to haul out. The drains can be closed after this point only if the fresh water source (hose water) is deemed clean enough to be used to fill the pool. If the pool becomes too dirty or the water level cannot be kept within 8-12" of the deck, the pool must be drained so the seals have access to a dry resting area. The seals will then be kept cool with shade and if appropriate, by a source of running freshwater.

A back-up generator supplies power to the main building, including the Vet Lab, and to 1 electrical outlet in the Fish House. Opening the freezers and refrigerators should be kept to a minimum to help these units maintain temperature during power outages.

***Potential Infectious Disease Outbreaks***

If one of the seals held in captive care exhibit signs of ill health and an infectious agent is the possible cause, the seal will be immediately moved to the designated isolation area. If it is unknown whether an infectious agent is the cause, the seal will be isolated and assumed infectious as a precautionary measure. Immediate sampling will be conducted following the Sampling Protocol (the “Re-check exam sampling protocol”) and every effort should be made to identify the causative agent. Treatment will be started before a diagnosis is made based on likely etiology as determined by the veterinary team.

- The NMFS Quarantine Policy will be followed.
- All equipment (feeding and cleaning supplies, handling gear, etc.) used to care for potentially infectious seals will be marked with a solid black triangle and will not be used to care for healthy seals. Isolation equipment will be stored separately and will not re-enter the clean animal area unless properly sanitized, if at all.
- Other seals exposed to a potentially infectious seal will remain separate from all unexposed seals (i.e. seals held in the same enclosure as the seal that became ill will not be mixed with those in a separate enclosure).
- There will be 1 animal care crew to care for the ill seals and another to care for healthy seals. Personnel caring for ill seals should not enter the clean seal area until showering and changing clothes.
- In each of the following locations, it is unlikely that a separate food preparation area will be available for the isolation area. If this is the case, a “one-way” fish kitchen will be set up in which contaminated items never enter the fish kitchen or healthy seal area. (For example, the amount of fish will be weighed out, thawed, and placed in a bag that will be handed to the sick animal caretaker at the base of the stairs to tanks 1 and 2.)
- The isolated seal will not be returned to its original holding tank until lab results verify the seal is healthy and the disease non-contagious.

Northwestern Hawaiian Islands: In the NWHI, the isolation area will be a separate pen erected off of the beach to prevent transmission of disease to other captive and wild seals. For example, on French Frigate Shoals, a quarantine pen will be erected at the southwest corner of the barracks on Tern Island if necessary.

Aboard the transport vessel: During vessel transport between the NWHI and Oahu, a separate pen will be erected on a lower deck of the ship if available or a separate area where cross-contamination is least likely (such as the bow versus the stern or the port and starboard sides). There will be sufficient space, more than 20 feet, and a solid barrier between the healthy animal and isolation areas to prevent direct contact, including splashed pool water and airborne disease transmission. If possible, there should be a separate footpath to the isolation area.

Kewalo Research Facility: See the Hawaiian Monk Seal Captive Care Protocol for Kewalo Research Facility, 2007-2008.

